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Endophyte-free tall fescue pastures for small-scale dairy systems in the highlands of central Mexico

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ABSTRACT

The objective was to assess dairy cows in small-scale dairy systems grazing pastures of endophyte-free tall fescue (*Lolium arundinaceum* cv. Cajun II), or perennial ryegrass (*Lolium perenne* cv. Tetragrain), both associated with white clover (*Trifolium repens*). An on-farm double cross-over experiment was undertaken with 10 Holstein cows, continuously grazing 10 h/day of Cajun II or Tetragrain pastures, plus 4.6 kg DM of a concentrate/cow/day. Animal variables were milk yield, milk fat and protein content, live-weight, body condition score, and milk urea nitrogen. The analyses of pasture variables was with a split-plot experimental design. Pasture variables were sward height, net herbage accumulation, and chemical composition of herbage for CP, NDF, ADF, *in vitro* digestibility of organic matter, and estimated herbage intake from utilised metabolizable energy. There were significant differences between treatments for OM, ADF, IVDOM, and eME, with tall fescue showing higher IVDOM which resulted in a slightly higher eME value. An economic analysis compared incomes and feeding costs. There were no statistical differences in animal or pasture variables. Cajun II endophyte-free tall fescue pasture performed similarly to Tetragrain perennial ryegrass in animal, pasture, and economic variables.

Keywords: Intensive grazing, *Lolium arundinaceum*, *Lolium perenne*, Margin over feed costs, Small-scale dairy systems

Small-scale dairy systems (SSDS) are a key activity for the economic, nutritional, social and territorial development of rural communities all over the world (Hemme and Otte, 2010), to assure family livelihoods, and an option to overcome rural poverty (Espinoza-Ortega *et al.* 2007).

SSDS face difficult scenarios for their viabilities since milk prices have remained static but production costs constantly increase, jeopardizing the financial viability of small dairy farms (Prospero-Bernal *et al.* 2017). These systems have the economic scale as the weakest component in the assessment of their sustainability due to high feeding costs that limit the profitability and efficiency of farms (Prospero-Bernal *et al.* 2017). Concentrates represent up to 70% of total production costs (Heredia-Nava *et al.* 2017).

Therefore, there is a need for lower cost feeding strategies that increase the sustainability and competitivity of small dairy farms. An alternative is grazing pasture that has proven lower feeding costs, increased profitability, and enhanced sustainability (Prospero-Bernal *et al.* 2017).

However, limitations to grazing pastures in small-scale dairy farms are limited irrigation and high evapotranspiration during the dry season, coupled with high stocking rates.

These factors reduce the persistency of pastures based on perennial ryegrass (*Lolium perenne*), the choice species for temperate pastures given its high nutritive value; but not tolerant to water deficit. Tall fescue (*Lolium arundinaceum*) is a hardy temperate perennial grass species, which although of lower nutritive value, is more tolerant to water deficit, agroecological and management stresses.

A problem with tall fescue is that old varieties are infected with the endo-phyte fungus *Neotyphodium coenophialum*, which causes tall fescue texicosis in cattle that affects production and reproductive performance (Waller, 2009). Endophyte-free tall fescue varieties overcome these problems.

Therefore, the objective was to assess dairy cows in small-scale dairy systems grazing pastures of endophyte-free tall fescue (*Lolium arundinaceum* cv. Cajun II), or perennial ryegrass (*Lolium perenne* cv. Tetragrain), both associated with white clover (*Trifolium repens*)

MATERIALS AND METHODS

Study area: The experiment was conducted in the municipality of Aculco, State of Mexico, in the central highlands of the country, located between 20° 00′ and 20° 17′ N and 99° 40′ and 100° 00′ W, at a mean altitude of 2,440 m. It has a sub-humid temperate climate with rains in summer (mid-May to mid-October), and a dry season during winter and spring (mid-November to mid-May), with frosts between November and February. Mean annual temperature is 13.2°C, and 800 to 1,000 mm rainfall (Jaimez-García *et al.* 2017).

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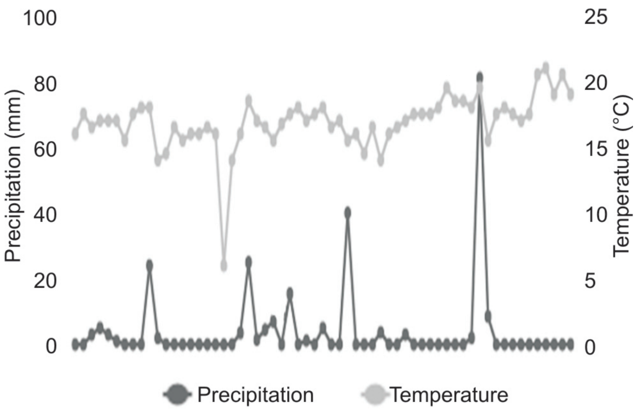

Fig. 1. Temperature and rainfall from 9 April to 3 June 2019 in the study area.

Work followed a participatory livestock research approach (Conroy 2005), through an on-farm experiment with a family of four participating small-scale dairy farmers who manage their land as a single farm. The experiment took place in the spring (dry season) from 9 April to 3 June 2018. Fig. 1 shows climatological data for temperature and rainfall during the experiment.

Animal variables: The experiment was a double cross-over design with 10 Holstein cows, selected from the farmers' herds. Cows were paired according to days in milk and milk yield on the week previous to the experiment, and members of each pair randomly assigned to treatment sequence. Pre-experimental variables were 14.3±6.2 milk/cow/day, 104±82 days in milk, and 473±56 kg live weight, with no statistical differences (P>0.05) between treatment sequences for these variables.

Experimental periods were 14 day duration each, with the first 10 d for adaptation to diet, and the last 4 d for recording and sampling. Fourteen days experimental periods (and shorter periods) are accepted when changes in feeding of dairy cows are not drastic, as has been well established by the INRA-Rennes group in France (Pérez-Prieto *et al.* 2011, Pérez-Ramírez *et al.* 2012).

Milk yield was recorded for four consecutive days with a clock spring balance (Thor brand made in Mexico with no model number) at the end of each experimental period and expressed as kg milk/cow/day. Individual milk samples per cow and per milking were taken, and milk and protein fat determined with an ultrasound milk analyser. Samples taken same way were for milk-urea-nitrogen determination by the enzymatic colorimetric method described by Chaney and Marbach (1962).

Live weight was recorded at the beginning of the experiment and at the end of each experimental period, by means of an electronic (Gallagher) weigh bridge. At the same time, body condition score (BCS) was determined on a 1(very thin) to 5 (very fat) point scale following Edmondson *et al.* (1989). The score reflects the amount of subcutaneous fat over the transverse apophyses of the lumbar vertebrae and around the base of the tail.

Herbage intake was estimated indirectly from utilized metabolizable energy following Pulido and Leaver (2001).

Pastures: The experiment took place on two pastures of 1.11 ha each; established in October 2017, in plots of volcanic acid soils (*p*H 5.7) with low contents of total Nitrogen (0.08%), and very low Phosphorus (2.44 mg/kg). Pasture 1 was established with tall fescue (*Lolium arundinaceum* cv. Cajun II). Cajun II is an endophyte free variety selected for improved nutritive value, tolerant to water deficit and high temperatures that has proven high yields, good nutritive value, and high persistency in sub-tropical Australia (Callow *et al.* 2003).

Pasture 2 was established with perennial ryegrass (*Lolium perenne* cv. Tetragrain) a tetraploid variety with high digestibility, tolerant to a range of acid and alkaline soils (*p*H 5.5 and 7.8). Tetragrain ryegrass has optimal growth at temperatures around 18°C, but reduces growth at 25°C, and requires constant irrigation as it is not tolerant to water deficit. Tetraploid ryegrasses have proven to be high yielding, and with higher nutritive value than diploid ryegrasses, with better results when associated with white clover (Guy *et al.* 2018).

Both pastures were associated with white clover (*Trifolium repens* cv. Fiona), sown immediately after the grass seed.

Seed rate for both pastures was 22 kg grass seed/ha plus 3 kg/ha of white clover. Due to a delay in available irrigation in Pasture 1 that precluded germination and emergence, it was reseeded with 17 kg/ha more of the Cajun II tall fescue seed. There was no need to reseed white clover as it establishes well in the area and surviving plants regenerated and rapidly colonised the pasture.

As is usual during the dry season, pastures were irrigated approximately every 28 days.

Fertilization on the same day as sowing was 80 N–80 P –60 K/ha, and pastures were thereafter fertilised every 28 days with 50 kg N/ha. Stocking rate was 4.5 cows/ha, with cows in each treatment sequence grazing each pasture alternatively every 14 day experimental period.

Treatments: Treatments were T×TF = continuous grazing of tall fescue pasture for 10 h/d (9: 00 to 19: 00 h), and T×PR = continuous grazing of perennial ryegrass pasture for 10 h/d (9: 00 to 19: 00 h). All cows additionally received 4.6 kg/cow/day of a commercial concentrate with 180 g/kg CP, divided in two equal meals provided at milkings.

Formulation of the commercial concentrate as stated in the label was rolled maize grain, cotton seeds, oleaginous seed meals, protected fat, sugarcane molasses, non-protein nitrogen, and minerals. The proportion of each ingredient was not specified on the label.

Pasture variables: Recording periods at pastures were similar to experimental periods, with variables recorded every 14 days. Pastures were nominally divided in two plots as replicates for statistical analysis. Sward height was from 40 measurements per pasture (20 for each nominal plot) with a rising plate grass metre following a 'W' pattern (Heredia-Nava *et al.* 2017).

Net herbage accumulation was estimated from 12 grazing exclusion cages (0.50 m × 0.50 m), six for each nominal plot, randomly distributed on each experimental period. Herbage was cut to ground level with handheld shears within a 0.16 m^2 quadrant outside the cage at the beginning of each experimental period (Day 0) and within the cage (Day 14).

Herbage collected was dried to constant weight at 60°C for 48 h in a draught oven for dry matter (DM) determination; and NHA estimated by difference and expressed as kg DM/ha for each period. NHA was determined following the method of López-González *et al.* (2015) with the following equation:

$$NHA = DMf\text{-}DMi$$

where NHA, net herbage accumulation (kg DM/ha); DMi, initial mean DM weight of herbage outside of a cage at day 0 (kg DM/ha); DMf, final mean DM weight of herbage inside of the cage at day 14 (kg DM/ha).

Samples of hand-plucked herbage simulating grazing from each pasture were taken during the last day of each period, and composite concentrate samples were dried as described above, and ashed at 550°C in a muffle furnace to determine organic matter (OM) (Celis-Alvarez *et al.* 2016).

Herbage and concentrate samples were dried in a draught oven at 65° C till constant weight to determine dry mattr (DM), and ashed in a muffle furnace at 550° C to determine organic matter (OM). Analytical methods for the determination of chemical composition of feeds were those reported by Jaimez-García *et al.* (2017): the Kjeldahl method for crude protein (CP); neutral detergent fibre (NDF), acid detergent fibre (ADF), and *in vitro* digestibility of organic matter (IVDOM) by the micro-bag technique, Enzymatic *in vitro* organic matter digestibility (IVOMD) followed Riveros and Argamentaría (1987). Estimated Metabolizable (eME) of feeds was calculated from IVDOM with the AFRC (1993) equation.

Economic analysis: Partial budget analysis was performed (Dillon and Hardaker, 1980), taking into account only feeding costs represented by supplemented concentrate and pasture costs (amortization of establishment costs, irrigation, and fertilization), and incomes from the sale of milk; expressed in US dollars.

Statistical analyses: Pasture variables were analysed with a split plot design by ANOVA where treatments (grass species in pastures) were main plots as fixed effects, and experimental periods as split-plots (random effects) (Kaps and Lamberson, 2004), with the following model:

$$Y_{ijkl} = \mu + r_i + T_j + E_k + p_l + Tp_{jl} + e_{ijk}$$

where, μ, General mean; r, Effect of replicates i = 1,2; T, Effect of pasture treatment (Main Plot) j = 1, 2; E, Error term for Main Plots [r (T)ij]; p, Effect of experimental periods (split–plot) k = 1, ..., 4; Tp, Interaction term between treatments and measurement periods; e, Error term for split plots.

The Tukey test was applied when significant differences were detected ($P \leq 0.05$).

Animal variables were analysed with a double cross-over design with 5 cows per treatment and 4 experimental periods following the treatment sequences T×TF – T×PR – T×TF – T×PR and T×PR – T×TF – T×PR – T×TF. Experimental periods were 14 days long, 10 days for adaptation and 4 days for recording.

The model for analyses of animal variables was:

$$Y_{ijkl} = \mu + S_i + C_j(_i) + P_k + T_l + e_{ijkl}$$

where μ, General mean; S, Treatment sequence (i= 1,2); C, Cows within sequence (j= 1, …, 5); P, Experimental periods (k=1, …4), T, Pasture treatments (1,2), and e, Residual error term.

RESULTS AND DISCUSSION

Table 1 shows results for animal variables where there were no significant differences ($P>0.05$) between pasture treatments. There were also no significant differences among periods ($P>0.05$), except for estimated herbage DM intake (HDMI) which was significantly higher ($P<0.05$) in Periods 1 and 2 compared with Periods 3 and 4.

Although there were no significant differences for animal variables, milk yield showed a pattern that followed the significant ($P<0.05$) changes observed in estimated herbage intake; with milk yields decreasing in time as estimated herbage intake decreased. This is similar to observations by Hernández-Mendo and Leaver (2006), who mentioned

Table 1. Animal variables

Variable	Treatment		SEM	Experimental period				SEM
	T×TF	T×PR		P1	P2	P3	P4	
MY (kg/cow/d)	13.3	13.5	0.55^{NS}	15.8	14.0	12.4	11.6	0.55^{NS}
Milk fat (g/kg)	36.3	36.1	1.30^{NS}	34.7	37.9	38.8	33.3	1.30^{NS}
Protein (g/kg)	31.1	30.2	0.58^{NS}	30.4	31.8	31.3	29.1	0.58^{NS}
MUN (mg/dl)	8.0	8.4	0.90^{NS}	6.1	6.5	9.0	11.0	0.90^{NS}
LW (kg)	466.0	471.0	4.75^{NS}	483.0	470.0	459.0	463.0	4.75^{NS}
BCS (1–5)	2.4	2.4	0.05^{NS}	2.6	2.4	2.2	2.4	0.05^{NS}
HDMI (kg DM/cow/d	6.3	6.7	0.30^{NS}	7.8^a	7.1^a	5.6^b	5.5^b	0.30*

T×TF, Continuous grazing of *Lolium arundinaceaum;* T×PR, Continuous grazing of *Lolium perenne;* MY, Milk yield; MUN, Milk urea nitrogen; LW, Live weight; BCS, Body condition score; HDMI, Estimated herbage DM intake; SEM, Standard error of the mean; NS ($P>0.05$); *($P<0.05$).

Table 2. Sward height and net herbage accumulation (NHA)

Variable	Treatment		SEMTX	Experimental period				SEM
	T×TF	T×PR		P1	P2	P3	P4	
Sward height (cm)	5.6	5.6	0.18[NS]	7.3[a]	4.1[b]	5.5[a]	5.5[a]	1.42*
NHA (kg DM/ha)	510.0	500.0	7.17[NS]	624.0	399.9	485.5	511.0	92.46[NS]
NHA (kg DM/ha/d)	36.3	35.7	0.40[NS]	44.5	28.6	34.7	36.4	6.55[NS]

T×TF, Continuous grazing of *Lolium arundinaceaum;* T×PR, Continuous grazing of *Lolium perenne;* SEMTX, Standard error of the mean for treatments (main plots); SEMex, Standard error of the mean for experimental periods (split-plots 4, NHA= Net herbage accumulation, NS= (P>0.05); *(P<0.05).

the close relationship linked to DM and energy intake. In the current experiment, estimated herbage intake reflected variations in milk yields.

Results are lower than reports by Macoon *et al.* (2011), who reported daily milk yields of 16.5 kg/cow under grazing with supplements. Results, however, are similar to previous reports in the study area (Martínez-García *et al.* 2015), who reported milk yields of 13 kg/cow/day.

Table 2 shows results for pasture variables. There were no significant differences (P>0.05) for sward height between pasture treatments, although there was a significant difference among periods (P<0.05) with a sharp decline from P1 to P2, to recover slightly in the last two periods. Mean grass metre sward height was above the 5 cm threshold under which difficult grazing conditions may limit herbage intake. Mayne *et al.* (2000) established that the optimal sward height for continues grazing is between 5.0 and 8.0 cm. Under 5 cm there are limitations to herbage intake by grazing cattle.

Mean NHA was 505 kg DM/ha/period, representing a daily growth of 36 kg DM/ha/d, with a mean herbage availability of 7.2 kg DM/cow/d.

Although grass metre sward heights may have been above the 5 cm threshold under which grazing conditions limit intake, herbage growth was only 7.2 kg DM/cow/d, limiting herbage intake to just a mean of 6.5 kg DM/cow/d representing a 90% grazing efficiency.

Low NHA was due to the lack of rain during the experiment as shown in Fig. 1 with just above 90 mm rain over the experiment, with rains starting till June. The limited irrigation available during the experiment was not enough to overcome the water deficit.

As live weight figures show, cows were small and live weight as is usual in these small-scale dairy systems, with low body condition score as is characteristic of Holstein cows, which tends to be lower in small-scale dairy systems given usual underfeeding conditions. Body condition scores were similar to reports of 2.3 by Celis-Alvarez *et al.* (2016).

Milk fat contained the requirement of 30 g/L established by Mexican standards. The forage to concentrate ratio of 60 : 40 enabled the observed milk fat contents. Bargo *et al.* (2002) reported a 13% milk fat content decrease in grazing cows when concentrate supplementation was above 5 kg DM concentrate/cow/d.

Observed milk fat content in the work herein reported are similar to reports from other countries like Roche *et al.* (2006) in New Zealand (35.9 g/kg) or Pérez-Prieto *et al.* (2011) in France (36.6 g/kg).

Herbage intake under grazing is determined by factors as herbage availability, sward height, herbage quality, the type and amount of supplements, and grazing pressure (Oliveira *et al.* 2007). Ample herbage allowance, above 20 kg DM/cow/d enables high herbage intakes (Bargo *et al.* 2002). Herbage allowance was low with just over 7.2 kg/cow/d.

The low NHA hampered herbage intake in spite of adequate grass metre sward height, not following statements by Pulido and Leaver (2001) who reported increased

Table 3. Chemical compositon of herbage and estimated metabolizable energy (eME) (MJ/kg DM)

Variable	Treatment		SEMTx	Experimental period				SEM
	T×TF	T×PR		P1	P2	P3	P4	
DM	252.2	205.9	32.78[NS]	214.1[b]	263.6[a]	218.1[a]	220.4[a]	21.19*
OM	869.5	834.2	24.92*	863.4	840.0	855.5	848.5	9.95[NS]
CP	175.3	215.2	28.24[NS]	169.5[b]	174.0[a]	227.7[a]	209.8[a]	28.18*
NDF	471.8	452.0	13.99[NS]	455.2[a]	481.9[a]	467.3[a]	443.1[b]	16.62*
ADF	230.3	215.2	6.73*	225.5[a]	241.9[a]	215.3[a]	208.4[b]	14.54*
IVDOM	639.7	617.6	10.56*	640.3[a]	594.3[b]	655.0[a]	624.9[a]	25.98*
Eme	10.0	9.7	0.25*	10.0[a]	9.3[b]	10.3[a]	9.8[a]	0.41*

T×TF, Continuous grazing of *Lolium arundinaceaum;* T×PR, Continuous grazing of *Lolium perenne;* SEMTx, Standard error of the mean for treatments (main plots); SEMex, Standard error of the mean for experimental periods (split-plots 4; DM, Dry matter; OM, Organic matter; ; CP, Crude protein; NDF, Neutral detergent fibre; ADF, Acid detergent fibre; IVDOM, *In vitro* digestibility of organic matter; eME, Estimated metabolizable energy; NS, (P>0.05); *(P<0.05).

herbage intake, and increased milk yields, with increasing sward heights. Dalley *et al.* (1999), Bargo *et al.* (2002) and Pérez-Prieto *et al.* (2011) stated that pasture intake of cows is restricted at herbage availabilities under 20 kg DM/cow/day.

Table 3 shows the chemical composition of the simulated grazing samples of herbage. There were significant differences (P<0.05) between treatments for OM, ADF, IVDOM, and eME, with tall fescue showing higher IVDOM which resulted in a slightly higher eME value. There were significant differences (P<0.05) among periods for all variables except OM.

Increased protein content with time may have been due to improved pasture condition from rains in May (Fig. 1) enhancing herbage regrowth as observed in the significant (P<0.05) increase in sward height (Table 2). Also, short pastures have a higher proportion of protein rich leaves in comparison with stems.

Herbage quality in both pastures was high, with over 180 g CP/kg DM, and between 400 g and 500 g NDF/kg DM, with high eME; both sustaining moderate milk yields in spite of restrictions to herbage intake due to low NHA.

Table 4 shows the partial budget analysis for feeding costs and returns from milk sales expressed in US dollars.

Given the slightly higher cost of tall fescue seed, there is a minor non-significant advantage for the perennial ryegrass pasture.

The economic analysis showed a marginal advantage of the Tetragrain perennial ryegrass pasture compared to the endophyte-free Cajun II tall fescue, given the higher price of seed, and therefore higher amortization of establishment costs, and the numerically lower milk yield.

Both pasture treatments showed similar profit margins expressed as the margin over feeding cost ratio, that for both pastures was above US$ 2.

An important aspect to note is the cost of concentrate per kg DM that was 8.5 times more expensive than the cost per kg DM of pasture, representing 85% of feeding costs.

Table 4. Economic analyses

	T×TF	T×PR
Feeding costs		
Concentrate (US$)	425.040	425.040
Pasture (US$)	74.080	71.280
Total (US$)	499.120	496.320
Feeding cost per cow (US$/cow)	99.820	99.260
Daily feeding cost per cow (US$/cow/d)	1.780	1.770
Feeding cost per kg (US$/kg milk)	0.134	0.131
Incomes		
Milk sales (USD$)	1,005.480	1,020.600
Margins over feeding costs		
Total margins (US$)	506.360	524.280
Margin per cow (US$/cow)	101.270	104.850
Margin per day (US$/d)	9.040	9.360
Margin per kg milk (US$/kg milk)	0.135	0.138
Income over feeding costs ratio (US$)	2.010	2.050

Prospero-Bernal *et al.* (2017) identified the economic scale as the weak point in the sustainability of these small-scale dairy systems, where feeding costs were identified as areas of opportunity given that concentrates comprise a large percentage of those costs.

Results showed that Cajun II endophyte-free tall fescue pasture under grazing performed similarly to the Tetragrain perennial ryegrass pasture in terms of animal performance, herbage growth and chemical composition, and in the economic analyses. However, since tall fescue is more tolerant to water deficit conditions, results encourage further research on the role that tall fescue may play under grazing in the feeding strategies of small-scale dairy systems.

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