



Evaluation of energy supplements in dual purpose cows in a silvopastoral system

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Received: 3 December 2022; Accepted: 22 February 2023

ABSTRACT

The objective was to evaluate the effect of three energy concentrates on dietary nitrogen utilisation efficiency, milk production and milk composition. Twelve dual-purpose cows fed in a silvopastoral (SSP) system with *Leucaena leucocephala* and *Digitaria swazilandensis* were fed four treatments, supplemented with 25 MJ/cow/day of sorghum, molasses and gluconeogenic precursors, plus the control treatment, using a 4×4 Latin square experimental design with three replicates and experimental periods of 14 days and a total duration of 56 days. No significant differences were found for milk yield, milk chemical composition and milk urea nitrogen (MUN). No significant differences were found for cow live weight and dry matter intake. Forage analysis showed significant difference for crude protein (CP) concentration. There was no significant difference for average values of dry matter (DM) yield, neutral detergent fibre (NDF), acid detergent fibre (ADF), lignin and *in vitro* DM matter. For *Leucaena*, a significant difference was found only in DM yield, with period 4 being the highest yielding period compared to periods 1, 2 and 3. There was no significant difference for CP, NDF, ADF, lignin and *in vitro* DM digestibility. This work allows to understand the interaction of CP metabolism with DM. It is concluded that SSP provides sufficient dietary energy to cows in production for dual-purpose cows ranching.

Keywords: Energy concentrate, *Digitaria swazilandensis*, *Leucaena leucocephala*, Nitrogen efficiency, Silvopastoral system

Tropical grasses are difficult to digest due to their high fibre content because they contain 20-70% cellulose and net energy capture ranges between 10-35% of the energy consumed, also the crude protein (CP) content is low, which makes it necessary to supplement to cover the N requirements, since, to ensure proper functioning of the rumen, cattle require 12.8 g of N per kg DM in the diet (Gaviria *et al.* 2015). Studies by López-Vigoa *et al.* (2017) indicate that silvopastoral systems (SSP) provide a diet equivalent to 11-16% CP and high forage availability for cattle, allowing milk yields between 3,000 and 16,000 kg/ha/year. SSP with *Leucaena leucocephala* are related to a diet with higher protein content and lower neutral detergent fibre (NDF) which favours an increase in animal productivity, however, an excessive intake of CP could

face high losses of N in urine and milk. For adequate utilization of CP, cattle need additional sources of energy; it is unknown if these diets are balanced or if it is necessary to use energy supplements, this makes it important to study the fermentative dynamics of forages (Gaviria *et al.* 2015). An alternative feeding method to balance diets in SSP is the use of additives to increase the intake of digestible energy and metabolizable energy with the aim of increasing the productivity of the system (Mendoza-Martínez *et al.* 2008). In tropical regions, it is supplemented with cereal grains such as sorghum (Schroeder *et al.* 2004), molasses which is also an excellent source of energy (Martín 2004) and currently there are biotechnological developments such as gluconeogenic precursors that some farmers are using (Leyva-Orasma *et al.* 2020). It is necessary to carry out research work on the study of energy supplements that balance the energy requirements and improve the utilisation of *Leucaena leucocephala* protein in SSP. Therefore, the objective of this study is to evaluate three energy supplements to determine their efficiency in the utilisation of nitrogen contained in the diet of dual-purpose cows fed in a SSP with *Leucaena leucocephala* and *Digitaria swazilandensis* grass.

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MATERIALS AND METHODS

Location of the experimental site: The present work was carried out in the municipality of La Concordia in the State of Chiapas, located in country south Mexico, with an average altitude of 550 m above sea level, with coordinates 15°41'N and 92°37'W. The climate is warm sub-humid with summer rains and semi-warm humid, with an annual rainfall of 1,450 mm (SEGOB 2010).

Grassland management and establishment: The ranch has 15 hectares established as SSP with *Leucaena leucocephala* and *Digitaria swazilandensis* grass, divided into 20 paddocks of 0.75 hectares for rotational grazing, with gravity irrigation support every 14 days in the dry season, the cows stay two days per area after which they have a recovery period of 40 days.

Development of the experiment: The experiment was carried out with 12 cows, with an average live weight of 456.1 kg and milk production of 7.84 kg/cow/day, in the second-third of lactation. The cows were accommodated using a statistical arrangement of 4×4 Latin square repeated three times. The experiment had a duration of 56 days, with experimental periods of 14 days; of which 10 days were for adaptation to the diet and four for sampling. The periods were established according to Miguel *et al.* (2014).

The sequence of treatments for the experiment was randomised and each cow within the square was also randomised to the sequence. The square 1 and 3 were randomised and the second was mirrored to the first square.

Treatments: The treatments evaluated were different energy supplements to complement the energy required by the cows, which were formulated to supply 25 MJ/cow/day of sorghum, molasses and gluconeogenic precursors, as well as the control treatment. The treatments were: T1= Molasses + continuous grazing of one SSP; T2= Sorghum + grazing of one SSP; T3= Gluconeogenic precursors + grazing on one SSP; T4= Control + grazing on one SSP. All cows had *ad lib.* access to water. The treatments were formulated considering that the silvopastoral system provides a good amount of crude protein (López-Vigoa *et al.* 2017), as they are deficient in energy (Gaviria *et al.* 2015).

Animal variables: Cows were milked once a day at 6:00 hours. Milk was individually weighed using a 31 kg milk weigher, also calves less than two months of age prior to weaning were weighed before and after they received suckling at the dam's foot after milking, to determine residual milk consumption, and both data were used to calculate milk yield. In addition, a milk sample was taken to determine its chemical composition (fat, protein and lactose), which was determined by ultrasound, using a Lactoscan equipment model Milkotronik Ltd., Nova Zagora, Bulgaria. A milk sample was also taken to determine milk urea nitrogen (MUN), using the colorimetric method (Chaney and Marbach 1962).

The live weight of the cows was recorded at the beginning and at the end of each experimental period, using a B&B

portable scale model 8100SS with a maximum capacity of 5000 kg.

Voluntary intake was estimated following the procedures of Hernández-Mendo and Leaver (2006) through the metabolisable energy used. Total DM intake was estimated indirectly from the energy requirements (AFRC 1993) of each cow and the ME concentration of the diets.

As the objective of the work was to measure milk yield, calves were not measured for milk intake during the experiment, however calves were weighed afterwards to estimate milk intake. The average milk intake was 1.4 kg of milk. This amount of milk is not reported in the results of the study. Mention is made in materials and methods, however, as they are indirectly estimated results, they are not considered as results and are only mentioned.

Samples from the *Leucaena leucocephala* and *Digitaria swazilandensis* pastures were taken at the beginning of each period to determine dry matter (DM) content by drying in an oven at 60°C for 48 h until reaching a constant weight (AOAC 1997). Crude protein (CP), acid detergent fibre (ADF), neutral detergent fibre (NDF) and lignin content were determined by near infrared spectroscopy (NIRS), using a Buchi NIR Flex N400 spectrometer (Büchi) (Cozzolino *et al.* 2006, Sánchez-Valdés *et al.* 2022).

Metabolisable energy was estimated using the CSIRO (2007) formula:

$$ME = 0.172 \text{ IVDMD (\%)} - 1.707$$

Where ME, metabolisable energy; IVDMD, *in vitro* dry matter digestibility (CSIRO 2007).

Experimental design: A 4×4 Latin square experimental design repeated three times was used (Kaps and Lamberson 2004). The animal production variables of the experiment were analysed using the statistical package Minitab 19, under the following mathematical model:

$$Y_{ijkl} = \mu + S_i + C_{j(i)} + P_k + t_l + e_{ijkl}$$

Where μ , Overall mean; S, effect due to squares i=1, 2, 3; C, effect due to cows within squares j, 1, 2, 3...12; P, Effect due to experimental period k, 1, 2, 3, 4; t, effect due to treatment l, 1, 2, 3, 4; and e, residual error term.

Forage production variables (*Digitaria swazilandensis* and *Leucaena*) were analyzed using a completely randomized design, using the package Minitab 19, with the following mathematical model:

$$Y_{ij} = \mu + t_i + e_{ij}$$

where μ , Overall mean; t, periods effect i,1,2,3,4; e, residual error term.

RESULTS AND DISCUSSION

Table 1 shows the results of yield and chemical composition of *Digitaria swazilandensis* by evaluation period, with no significant differences ($P > 0.05$) in the grass yield with an average value of 921.33 kg DM/ha. In the crude protein (CP) content of *Digitaria swazilandensis*, significant differences were observed ($P < 0.05$), with a higher crude protein content in periods 3 and 4 compared to periods 1 and 2, with a mean of 103.51 g CP/kg. The highest

Table 1. Yield and chemical composition of grass *Digitaria swazilandensis*

Variable	Period				Mean	SEM
	1	2	3	4		
Yield (kg DM/ha)	898.86	890.07	1226.73	669.66	921.33	155.38 ^{NS}
CP (g/kg DM)	85.54 ^a	86.94 ^a	96.78 ^a	144.79 ^b	103.51	6.83 [*]
NDF (g/kg DM)	676.14 ^b	562.83 ^a	581.16 ^a	578.11 ^a	599.56	11.92 [*]
ADF (g/kg DM)	423.20 ^a	343.13 ^b	400.21 ^a	382.48 ^a	516.34	8.53 [*]
Lignin (g/kg DM)	75.38	47.74	75.37 ^a	65.23	65.92	9.89 ^{NS}
IVDMD (g/kg DM)	726.04	731.04	728.67	721.78	726.88	6.93 ^{NS}

CP, crude protein; NDF, neutral detergent fibre; ADF, acid detergent fibre; IVDMD, *in vitro* digestibility matter dry; SEM, standard error of the mean; 1, 2, 3, 4, evaluation periods.

value being observed in period 4 with 144.79 g CP/kg, and the lowest in period 1 with 85.55 g CP/kg. The NDF and ADF content did not show significant differences ($P>0.05$) among the periods and the estimated metabolisable energy did not show significant differences ($P>0.05$), as did lignin (Table 1).

Table 2 shows the yield of *Leucaena leucocephala*, in which there are significant differences ($P<0.05$) among the periods evaluated; higher forage production was observed in period 4 compared to period 1. No significant differences ($P>0.05$) were observed in protein content (CP), with a mean value of 187.64 g CP/kg. Regarding the amount of structural carbohydrates, no significant differences ($P>0.05$) were observed for both lignin, NDF and ADF (Table 2). The *in vitro* digestibility of dry matter and estimated metabolisable energy showed no significant differences ($P>0.05$) between the evaluation periods (Table 2).

In terms of milk yield, no significant differences were

found ($P>0.05$), with an average milk yield of 9.64 kg/cow/day (Table 3). Table 3 shows the live weight of the cows, with a mean of 460.81 kg (± 3.89 kg), with no significant differences ($P>0.05$) among treatments. Regarding the chemical composition of the milk, there are no significant differences ($P>0.05$) in fat, lactose and protein content, the average figures found were 40.20 g/kg, 48.85 g/kg and 31.82 g/kg respectively (Table 3). According to the values found for milk urea nitrogen (MUN), there were no significant differences among treatments ($P>0.05$), the mean was 6.02 mg/dL (Table 3). There were no significant differences ($P>0.05$) in dry matter intake (DMI) among treatments, the mean value being 9.22 kg DM/cow/day as shown in Table 3.

Silvopastoral systems have the characteristic of providing a good crude protein content, however they are low in metabolisable energy, but in the present study it is observed that the control treatment has the same results as the treatments that were supplemented with energy, which

Table 2. Yield and chemical composition of *Leucaena leucocephala*

Variable	Period				Mean	SEM
	1	2	3	4		
Yield (kg DM/ha)	726.46 ^b	985.56 ^b	859.83 ^b	2613.39 ^a	1296.31	29.41 [*]
CP (g/kg DM)	217.20 ^a	177.52 ^b	189.59 ^b	166.25 ^b	187.64	2.55 [*]
NDF (g/kg DM)	483.95	442.26	496.58	433.94	464.18	26.60 ^{NS}
ADF (g/kg DM)	301.11	335.18	312.66	307.55	314.13	19.03 ^{NS}
Lignin (g/kg DM)	75.15	62.44	66.49	54.53	64.65	4.58 ^{NS}
IVDMD (g/kg DM)	719.43	733.58	735.49	730.33	729.70	9.34 ^{NS}

CP, crude protein; NDF, neutral detergent fibre; ADF, acid detergent fibre; IVDMD, *in vitro* digestibility matter dry; SEM, standard error of the mean.

Table 3. Productive response of cows to supplementation treatments and dry matter intakes

Variable	Treatment				Mean	SEM
	Control	Precursors	Molasses	Sorghum		
FCM 3.5% (k)	10.73	10.61	10.33	11.07	9.64	0.50
Live weight (kg)	456.92	464.49	464.25	457.58	460.81	3.73
Milk						
Fat (g/kg)	41.10	39.30	38.50	42.10	40.20	0.20
Protein (g/kg)	31.90	32.00	31.60	31.75	31.82	0.02
Lactose (g/kg)	49.03	49.04	48.88	48.43	48.85	0.03
MUN (mg/dL)	6.43	6.26	6.56	4.84	6.02	0.48
DMI (kg DM/cow)	8.87	9.45	9.33	9.46	9.22	0.32

FCM, fat corrected milk; MUN, milk urea nitrogen; DMI, dry matter intake; SEM, standard error of the mean.

indicates that the SSP system alone covers the requirements of the cows. A factor that could determine the yield of the cows is the substitution effect observed in the treatments, the dry matter intake (DMI) of the cows was similar in all treatments (Stockdale 2000), which mentions that the substitution effect explains the variation in milk yield, as well as the possible weight gain of the animals. Another important factor that may have influenced milk yield is the CP content provided by *Leucaena leucocephala* (Bottini-Luzardo *et al.* 2016). The milk yield results are within the production ranges for dual-purpose cows who grazed on SSP in the tropics, agreeing with Esparza-Jiménez *et al.* (2021) who reported that there is no significant difference in milk production in cows supplemented on a SSP, with a yield of 7.7 kg milk/cow/day, on the other hand Arjona-Alcocer *et al.* (2020) in a study where molasses, sorghum and citrus pulp were included as energy source reported milk yield results of 4.1 kg milk/cow/day. In both studies, lower milk yields were obtained.

One of the factors that influences the fat content of milk is the amount of forage consumed by cows (Wanapat *et al.* 2018, López-González *et al.* 2020), which is consistent with the results of this study, where the highest fat content is observed in treatment four, being one of the treatments in which there is a higher consumption of forage, with no significant differences ($P > 0.05$). In this regard Tinoco-Magaña *et al.* (2012), found no significant differences in fat content, on the other hand, Arjona-Alcocer *et al.* (2020), report a higher fat content in cows supplemented with different energy treatments. It is known that there is a directly proportional relationship between milk yield and the amount of fat in milk, according to data from INRA (2007), which mentions that 2 g/kg of fat is reduced with an increase of 10 kg of milk, which may explain why the fat content results of Arjona-Alcocer *et al.* (2020) are different from those found in this work, and are also different from the data reported by Barros *et al.* (2017) in Arlington, Wisconsin who found that in Holstein cows with 11.8% CP diets the fat levels were 43.4 g/kg, lactose 46.9 g/kg and protein 33.3 g/kg. It also differs with Celis-Alvarez *et al.* (2021) who reported values for fat as 38.2 and 38.8 g/kg and for protein as 29.03 and 28.79 g/kg in a small-scale milk production system in the central plains of Mexico.

The average dry matter intake by the animals was 9.22 kg/cow/day, which means 2% of the live weight of the animal, the low intakes by the animal can be explained by the amount of structural carbohydrates contained in both *Leucaena leucocephala* and *Digitaria swazilandensis*, which are two factors that limit the voluntary intake by the animal (Mayne *et al.* 2000), mentioning that forage quality influences the voluntary intake. The results of this work are similar to those reported by Pardo *et al.* (2008), who reported average intake values of 7.4 kg DM/animal/day.

The amount of milk urea nitrogen (MUN) is an indicator of the nutritional status of cows and their lactation (Mitchell *et al.* 2005), mentioning that adequate levels of

urea nitrogen in milk are between 10 and 16 mg/dL for a typical lactation. In this study, the results of urea nitrogen in milk are below the mentioned parameters (6.02 mg/dL on average), which indicates that the crude protein content of both *Leucaena* and *Digitaria* are relatively low and that supplementation may be required. On the other hand, it is mentioned that an excess of dietary protein has implications for both the environment and the reproductive performance of the animals. The results of milk urea nitrogen are similar to those reported by Pardo *et al.* (2008), who reported average values of 7.3 mg/dL. On the other hand, Kohn *et al.* (2002) reported results of NUL and found that in 13.1% CP diets the values were 6.8 and 9.5 mg/dL, different from those reported in this work.

It is concluded that the SSP provides sufficient dietary energy to cows in production characteristic of dual-purpose farms in the tropics, since the addition of energy in the diet of cows in SSP does not significantly affect milk production.

ACKNOWLEDGEMENTS

The authors thank the producer and his family, as well as all the people who made this experiment possible with their participation. We omit their names to guarantee their safety. Thanks to the National Council of Science and Technology (CONACYT) for the scholarship awarded to Joel Galvez Luis for his postgraduate studies.

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