Paper ID: 614
Theme: 2. Grassland production and utilization
Sub-theme: 2.1. Quality, production, conservation and utilization

The effects of *Tithonia diversifolia* on dairy cow performance

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Keywords: Forage, Supplementation, *Tithonia diversifolia*

Introduction

Southeast, South and Central West are the main milk producing regions in the Brazil. Especially in the states of Minas Gerais, Goias and Sao Paulo, the tropical climate is very characteristic, with hot and rainy summers, and dry winters. Dry winters in these states are characterized by scarcity of pasture herbage mass, which directly influence the volume of milk produced.

The high volume of milk produced in summer and low volume of milk produced in winter (*i.e.* seasonality of production which is about 20% of total milk volume) directly affects dairy farmers by reducing its revenue during dry winters due to a drop in milk yield. In addition, it increases the production costs by offering additional roughage supplements to the cattle (sugarcane fresh plus urea, corn silage or sorghum silage), or by feeding more concentrates and/or greater labour costs.

Research evaluating the potential of the *Tithonia diversifolia* in improving milk yield and quality is extremely limited. This research project seeks to develop tools to understand the potential impact on milk composition and cow performance and to evaluate the significance of its outcomes and to aid in the ongoing development of innovative approaches.

The aim of this study is to determine the effects of replacing up to 9.1% of sugarcane fresh and up to 6.3% of concentrates (DM basis) with *Tithonia diversifolia* fresh fed to lactating dairy cows. It is hypothesized that the initial replacement of a portion of the sugarcane fresh and concentrates (corn grain and soybean meal) with the *Tithonia diversifolia* fresh would not reduce dairy cow performance.

Materials and Methods

Animals and experimental design*:* The experiment was conducted as a crossover design with 3 groups of 3 cows, 3 treatments and three 21-d periods. The first 14 d of each period was used to gradually adapt animals to the diet, and the last 7 d was used for measurements. Nine lactating cross-bred Holstein-Zebu dairy cows (519 ± 53.3 kg of body weight and 66 ± 13.3 days in milk) were paired by milk yield and body weight, and randomly assign to 3 treatments. The experimental diets were formulated for crude protein (CP) and ether extract (EE) contents of 18% of DM and 1.4% of DM, respectively.

Feed intake*:* Quantities of feed offered and refusal were weighed daily (n=6), on an individual basis, for the determination of dry matter intake. Samples of the offered and refused feeds were collected daily and dried at 100 °C for 24 h for the determination of DM content. For chemical analysis, individual sample for each ingredient was pooled to make one composite sample per period.

Milk yield and composition: Cows were milked twice daily at 0700 and 1400 h, and milk was sampled during the a.m. and p.m. milkings on 3 consecutive days (d 17 to 19) in each period. Milk samples were preserved with a preservative (Bromopol tablet; D & F Control Systems Inc., San Ramon, CA) and stored at 4°C until sent to EMBRAPA-CNPGL (Dairy Research Centre, Juiz de Fora, MG, Brazil) for analysis of fat, protein, and lactose (AOAC, 1995) using an infrared analyzer (Bentley 2000, Bentley Instruments). Milk composition was corrected for differences in milk volume between a.m. and p.m. milkings. Milk urea–N (MUN) was analysed by colorimetry using a commercial kit (Sigma Diagnostics, St. Louis, MO).

Blood metabolites: On day 5 of the measurement period, at 2 and 6 h after the daily feed delivery, blood was withdrawn from the mammary vein of all cows from each treatment group. Samples were collected using 4 mL vacutainer tubes (13×75 mm; BD vacutainer - Plus blood collection tubes Ref 368521— Becton Dickinson, Belliver Industrial Estate, Plymouth, UK). The tubes were immediately transferred to the laboratory and the supernatant (i.e., serum) was centrifuged (1800 ×g, 20 min, +4 °C) and harvested for later analyses of glucose, urea, triglycerides, cholesterol, β-hydroxybutyrate (BHBA) and non-esterified fatty acids (NEFA).

Chemical analyses: Dry matter content of feeds samples was determined by oven-drying at 100 °C for 24 h (method 967.03, AOAC, 1995). Ash content was determined after 2 h of incineration at 600 °C in a muffle furnace (method 942; AOAC, 1995). Neutral detergent fibre (NDF) content was determined as described by Van Soest *et al*. (1991) without the use of sodium sulfite or heat stable α-amylase, and acid detergent fibre (ADF) following the method 973.18 of AOAC (1995). Concentration of total N was determined by Kjeldahl (Vapodest 20S, Gerhardt Instruments, Germany) and crude protein (CP) concentration was calculated as N×6.25. Ether extract (EE) content was determined by extraction with petroleum ether (method 920.39; AOAC, 1995) using a Soxtherm Fat Extractor (Gerhardt Instruments, Germany). Non-fibrous carbohydrate (NFC, % of DM) was calculated as 100− (CP + NDF + EE + Ash).

Statistical evaluation of experimental design*:* Data were analyzed in a cross over design using the proc MIXED procedure of SAS (SAS Inst. Inc., Cary, NC). Degrees of freedom were adjusted using the Kenward-Roger option. Means were compared using the LSMEANS/DIFF with treatments and period as fixed terms and cow nested within a period and within a sequence as a random effect. Day were analysed as a repeated measure. Different covariance structures for repeated measures analysis were tested for minimum values of Akaike's information criterion. Treatment differences were declared at $P \leq 0.05$.

Results and Discussion

The proportion of each ingredient and chemical composition of dietary treatments are shown in Table 1.

Table 1 - Ingredients and chemical composition (% DM) of diets (n = 3)

		Diets			
Item		Control	6.4%T	15.3%T	
Ingredients, % dry matter basis					
	Tithonia fresh	0.0	6.5	15.4	
	Sugarcane fresh	43.8	40.1	34.7	
	Corn grain coarse grind	31.2	30.2	29.1	
	Soybean meal finely ground	24.2	22.4	20.0	
	Limestone	0.79	0.79	0.79	
Dry matter and chemical composition (% in the DM)					
	Dry matter, %	63.8	61.6	59.0	
	Crude protein	18.5	18.4	18.4	
	Neutral detergent fibre (NDF)	29.4	30.6	32.2	
	Acid detergent fibre	15.1	16.4	18.2	
	Ether extract (EE)	1.4	1.4	1.4	
	NFC	46.1	44.8	42.9	
	Ash	4.5	4.8	5.1	
Abbreviations: T, *Tithonia diversifolia*; NFC, non-fibrous carbohydrates [NFC = 100 − (CP + NDF + EE + ash)].					

Dietary Tithonia treatments did not affected ($P \geq 0.48$) DM intake, milk yield, or milk composition (Table 2) compared to control. The DM intake, milk yield, or milk composition reported for cows in this study were typical of Brazilian cross bred dairy cows fed diets with high proportion of concentrates and fresh sugarcane (Cordeiro *et al*., 2007).

Table 2 - Intake, milk yield and composition of cow supplemented with 0, 6.4 or 15.3% of Tithonia fresh replacing sugarcane fresh and concentrates in a total mixed ration diet

	Control	6.4%T	15.3%T	SEM	*P*-value
Dry matter intake, kg/d	18.6	18.9	18.7	0.63	0.96
Milk yield, kg/d	22.7	23.1	22.8	1.49	0.98
Milk fat, %	3.58	3.48	3.45	0.127	0.76
Milk protein, %	3.23	3.15	3.08	0.082	0.48
Milk lactose, %	4.34	4.42	4.36	0.106	0.84
Milk urea-N, %	13.8	13.5	12.9	0.83	0.74
Abbreviation: SEM, standard error of the means.					

Glucose, urea, triglycerides, and cholesterol serum concentrations were not affected ($P \geq 0.39$) by additive treatments averaging 40.6, 28.4, 10.4 and 103.8 mg/dL, respectively (Table 3). Also β-hydroxybutyrate serum concentration was not affected (P=0.64) by inclusion of fresh Tithonia in the diet. However cows fed 15.3% of fresh Tithonia had lower (P<0.001) serum concentration of non-esterified fatty acids compared to other treatments at 6h after feeding.

Table 3 - Blood parameters of cow supplemented with 0, 6.4 or 15.3% of Tithonia fresh replacing sugarcane fresh and concentrates in a total mixed ration diet

						P-value	
	Control	6.4%T	15.3%T	SEM	Treatment	Time	Treatment × Time
Glucose, mg/dL	39.8	41.5	40.4	1.03	0.50	<0.0001	0.14
Urea, mg/dL	27.9	30.2	27.1	1.89	0.50	<0.0001	0.13
Triglycerides, mg/dL	9.9	10.1	11.2	0.7	0.39	0.33	0.43
Cholesterol, mg/dL	95.2	102.9	113.2	9.06	0.39	0.55	0.20
BHBA, mmol/L	0.89	0.68	0.81	0.161	0.64	0.74	0.89
NEFA, mmol/L	0.034	0.032	0.028	0.004	0.47	<0.0001	0.001
Abbreviations: BHBA, β-hydroxybutyrate;NEFA, non-esterified fatty acids.							

Conclusion

Inclusion of Tithonia fresh up to 15.3% (DM basis) replacing sugarcane fresh and concentrates in a total mixed ration diet fed to dairy cows did not caused any change in intake, milk yield and composition. However, serum concentration of non-esterified fatty acids was lower in cows fed 15.3% of fresh Tithonia compared to other dietary treatments.

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Acknowledgement

The authors thank IFET (Barbacena-MG, Brazil) staff, including W Carvalho and R Pereira (animal care and sampling), and EMBRAPA-CNPGL staff, including B Carvalho (blood laboratory analysis). This study was funded by CAPES-PVE – Programa Ciência Sem Fronteiras (Brazil). Also we thank EMBRAPA-Rumen Gases, FAPEMIG-PPM, CNPq, CAPES-EMBRAPA, and DEPEB (UFSJ).