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Research paper

Lambs fed cassava silage with added tamarind residue: Silage quality, intake, digestibility, nitrogen balance, growth performance and carcass quality

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ABSTRACT

The objective of this study was to evaluate the effects adding tamarind residue to cassava silage on the quality of the silage and its *in vitro* ruminal fermentation, as well as the growth performance and carcass quality of lambs. A completely randomized design with four inclusion levels (0.00, 100, 200 and 300 g/kg) of tamarind residue (*Tamarindus indica* L.) added to cassava silage was used. Twenty mini-silos were used to prepare samples of the four treatments (five replicates each), which were opened after 56 days. Forty male lambs were fed cassava silage with added tamarind residue as the roughage and a concentrate (500:500 g/kg of feed) over 85 days. There was no effect of the tamarind residue-added cassava silage on effluent losses and dry matter (DM) recovery rates. However, there were linear and quadratic reductions in pH ($P < 0.001$) and the crude protein (CP) ($P < 0.001$), ether extract (EE) ($P < 0.001$) and ash ($P < 0.001$) contents and linear and quadratic increases in the DM, neutral detergent fiber (NDF) ($P < 0.001$), acid detergent fiber (ADF) ($P < 0.001$), non-fibrous carbohydrate (NFC) ($P < 0.001$), acid detergent lignin (ADL) ($P < 0.001$), and tannin ($P < 0.001$) contents. There was a linear increase in the maximum potential gas production from total carbohydrates and the gas production rate from NFCs ($P < 0.001$). The production rate of total gases ($P < 0.001$), lag phase duration ($P < 0.001$) and DM *in vitro* degradability ($P < 0.001$) were all reduced with the addition of tamarind residue in silage. There was no effect of the tamarind residue-added cassava silage on DM, CP, NFC or total digestible nutrient (TDN) intakes (g/d). However, there was a linear increase in the NDF intake ($P = 0.042$) and a linear reduction in the EE ($P = 0.038$) intake by lambs. There was an increase in the DM ($P < 0.001$), CP ($P < 0.001$), and NDF ($P < 0.001$) contents. There was linear effect on the DM ($P < 0.001$), CP ($P < 0.001$), and NDF ($P < 0.001$) contents and a positive quadratic on EE ($P = 0.018$) digestibility. There was linear

Abbreviations: ADF, acid detergent fiber; ADG, average daily gain; ADL, acid detergent lignin; CCW, cold carcass weight; CCY, cold carcass yield; CP, crude protein; DM, dry matter; DMI, dry matter intake; EE, ether extract; FCs, fibrous carbohydrates; GC, gastrointestinal tract contents; HCW, hot carcass weight; HCY, hot carcass yield; IVDMD, in vitro dry matter digestibility kinetics over 48 h; LDA, *longissimus dorsi* area; NDF_{ap}, neutral detergent fiber corrected for ash and protein; m_1, gas production rates from non fibrous carbohydrates; m_2, gas production rates from fibrous carbohydrates; mt, total gas production rate; NFCs, non-fibrous carbohydrates; TDNs, total digestible nutrients; vf_1, maximum potential gas production from non-fibrous carbohydrates; vf_2, maximum potential gas production from fibrous carbohydrates; vt, maximum potential gas production from total carbohydrates

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increase in nitrogen (N) fecal excretion ($P < 0.001$) and a quadratic decrease in N urinary ($P = 0.018$) excretion and N retention ($P < 0.001$). There was an increase in the hot and cold carcass weights and yields ($P < 0.001$) and in the *Longissimus dorsi* area (LDA) ($P < 0.001$) of the lambs. The addition of 300 g/kg tamarind residue to cassava silage is recommended because it reduces the production of gases and improves the DM content of the diet. In addition, this diet increases the DM *in vitro* digestibility and intake (DMI), N retention, hot and cold carcass yields, and LDA of lambs.

1. Introduction

Ensiling cassava (*Manihot esculenta*, Crantz.) is an important method of fodder preservation for maintaining the diets of animals during periods of shortage. Howeverthe low use of cassava silage in animal feed is related to the lack of knowledge of its nutritional levels and the actual potential of crop residues for use in the feeding and production of ruminants (Silva et al., 2015a; Polyorach et al., 2016). In addition the use of cassava silage is limited by its low dry matter (DM) content (220–250 g/kg) which causes difficulties in fermenting the silage (Santos et al. 2015). Another important factor relevant to the ensiling process is the high crude protein (CP) content of cassava (205 g/kg DM) which can lead to a buffering effect that hinders the reduction of the pH to levels optimal for silage fermentation (Fernandes et al., 2016; Ampapon et al., 2016). Thus residues originating from the agro-industry are under consideration for use as additives to silage forage plants that can increase DM content and reduce the risk of losses via undesirable fermentation (Gonzaga Neto et al., 2015 ; Oliveira et al., 2015a).

In this respectthe tamarind (*Tamarindus indica* L) a member of the Fabaceae family is one potential additive (Geron et al., 2015). This plant originates in Africa but can currently be found in many tropical countries. The pulp of its fruit is edible and the tamarind processing industry generates tamarind residues after the drying process (Wang et al., 2016). Therefore it was hypothesized that because of its chemical composition (DM = 880 g/kg feed and CP = 75 g/kg DM) dried tamarind residue could be added at up to 300 g/kg as an additive to improve cassava silage and reduce losses due to fermentation and that this silage could be used as a lamb diet where it would increase feed intake and digestibility and improve nitrogen (N) balance growth performance and carcass quality of lambs. The objective of this study was to determine the most effective level at which to add tamarind residue to cassava silage for improving the chemical composition and quality of the silage and to determine what effect this had on feed intake and degradability and on N balance growth performance and carcass quality of lambs fed this silage.

2. Materials and methods

This study was carried out in strict accordance with the recommendations in the Guide of the National Council of Brazil for the Control of Animal Experiments of the Federal University of Bahia, Bahia State, Brazil (Permit Number: 0002/140814).

2.1. Silage and treatments

The cassava used in the silage consisted of the above-ground biomass (manioc + leaf) of the *Recife* variety harvested after approximately six months of cultivation. Cassava silage was produced in an irrigated system and grown with 1.0 m between plants and 1.2 m between rows. After harvest and transport, the above-ground biomass was crushed into 3- to 4-cm pieces with the aid of sieves contained in a fodder machine. Tamarind residue was obtained from plants used in the tamarind pulp manufacturing industry, and it was dried in the sun for approximately 12 h to reach a DM content of approximately 850 g/kg. It was then ground into a meal using a forage machine with a 3-mm-diameter sieve.

After drying, the tamarind residue was mixed with the above-ground biomass of cassava to obtain a homogeneous mixture. To make the mini-silos (five replicates per treatment), polyvinyl chloride (PVC) tubes 100 mm in diameter and 50 cm in length (5-L capacity) were used. They had a Bunsen-type valve on the lid for the output of gases from the silo and a sand chamber for wastewater collection.

The treatments varied according to the levels of tamarind residue added (0.00, 100, 200 or 300 g/kg of feed) to the natural silage material. Cassava silage with tamarind residue added in the same proportions as prepared in the mini-silos was also prepared in barrels with a 200-L capacity (Poliembalagens®, São Paulo, Brazil). The material was compacted by trampling, and the barrels were sealed with a lid using a metal seal. The mini-silos were packed with the aid of a wooden plunger and filled to a density of 600 kg/m^3. Then, each pipe was closed with a PVC cap and sealed with plastic tape (Poliembalagens®, São Paulo, Brazil). The large barrels and mini-silos were stored in the shade at a temperature between 20 and 25 °C and were opened after 56 days.

After opening the mini-silos, the pH was measured with a pH meter (MA522 model, Marconi Laboratory Equipment, Piracicaba, Brazil). The effluent losses were quantified using the following equation proposed by Jobim et al. (2007): E (kg/t FM) = [(TWo − TWs)/(GMef)] × 100, where E = effluent yield (kg/t FMs), FMs = forage mass in the silage, TWo = total weight (silo + sand + sandbag) when the silos were opened (kg), TWs = total weight (silo + sand + sandbag) when the silos were sealed (kg), and GMef = green mass of the ensiled forage (kg). The gas losses were quantified using the following equation: GL (g/kg DM) = [(SWs − SWo)/(FMs × DMe)] × 100, where GL = gas losses during storage (g/kg of initial DM), SWs = silo weight during the ensilaging process, SWo = silo weight at opening, FMs = forage mass in the silage, and DMe = DM content of the forage in the

silage. The DM recovery rate in the silage was determined by the method proposed by Jobim et al. (2007) using the following equation: DMR (g/kg) = [(FMo × DMo)/(FMc × DMc)] × 100, where DMR = DM recovery rate, FMo = forage mass at silo opening, DMo = DM content at silo opening, FMc = forage mass at silo closing, and DMc = DM content of the forage at silo closing. The ammonia N (mg/g total N) content was determined as described by Bolsen et al. (1992). Samples of each mini-silo were dried in a forced-circulation oven at 55 °C for 72 h and subsequently milled in a knife mill with a 1-mm sieve.

2.2. In vitro trial using an in vitro gas production technique

For kinetic assays of degradation and gas production, an *in vitro* semiautomatic technique of measuring gas production was conducted according to Mauricio et al. (2003) and modified by Menezes et al. (2015). Samples were dried in an oven with forced ventilation at 55 °C for 72 h and ground in a mill with a 1-mm sieve; approximately 1 g of each sample of silage was incubated in glass jars (160 mL), and there were five replicates per treatment. To each vial, 90 mL of culture medium prepared according to Theodorou et al. (1994) was added manually; this medium had been previously reduced from aspersion with CO_2 to adjust pH to within the range of 6.8–7.0. In addition, 10 mL of ruminal fluid collected directly from the rumen of two cannulated sheep was added to each vial. From the start, ruminal fluid was homogenized and packed in thermoses previously heated with water at 39 °C and then filtered through a double layer of gauze. The buffer solution and the inoculum were added under CO_2 aspersion to ensure anaerobic conditions. Vials were immediately sealed with a rubber stopper, sealed and then kept in an incubator chamber at 39 °C. Pressure readings were performed at 2, 4, 6, 8, 9, 11, 12, 14, 17, 20, 24, 28, 34, 48, 72, 96 and 120 h.

Data obtained from the measurements of the cumulative production of gas were then subjected to the two-compartment model suggested by Schofield et al. (1994) as follows: $v(t) = [vf_1/(1 + e^{(2-4m1(lt))})] + [vf_2/(1 + e^{(2-4m2(lt))})]$, where v (t) is the maximum volume of the total gases produced, vf_1 represents the maximum gas volume fraction from the rapid degradation (from NFCs), vf_2 represents the maximum gas volume fraction from the slow degradation (from FCs), m_1 equals the specific growth rate for the fraction from the rapid degradation, m_2 is equivalent to the specific growth rate of the fraction from the slow degradation, lt is the lag time (or colonization time), and b (h^{-1}) and c ($h^{-0.5}$) are the constant fractional rates. The fractional rates (h^{-1}) combined with gas production (μ) were also calculated with the following equation: $\mu = b + c/Q_2$, wherein μ = the gas production rate (h^{-1}), and Q_2 is the duration of the initial digestion events (the lag phase) common to the two phases.

The fermentation residues were obtained by filtration in crucibles of porosity 1 (Pirex, Vidrotec, São Paulo, Brazil), which were dried for 48 h at 100 °C and weighed to calculate the DM degradability (DMD) values. The DMD was estimated after 48 h of fermentation.

2.3. Animal experiments and management

Forty non-castrated male lambs of undefined breed that were vaccinated and dewormed (six months old, 22.1 ± 3.00 kg BW (mean ± SD) were used. The experiment used a completely randomized design that had 4 treatments and 10 replicates.

The animals were housed in a covered shed in individual 1.61-m^2 pens equipped with drinking fountains, feeders and salt shakers. The experimental period lasted 85 days, with 13 days allowed for adaptation to the installation, location and diet. The residue inclusion levels were increased gradually, with three days at each level of inclusion, so that the animals that received treatments with a greater level of inclusion also initially received the lower levels of residue inclusion in the silage.

The diets were offered twice daily, at 9:00 and 16:00 h, in the form of a total mixed ration with roughage and concentrate (500:500 g/kg of DM). The cassava + tamarind residue mixture (0.00, 100, 200 or 300 g of residue/kg of DM included in the natural silage material) was used as roughage, and the concentrate was made from a cornmeal base with soybean meal. The provided feed and refused feed were weighed daily to monitor nutrient intake. Adjustments were made to ensure that the remains were between 100 and 200 g/kg DM. Water and mineral salts were provided *ad libitum*.

Diets were formulated according to the recommendations of the National Research Council (NRC, 2007) to meet the nutritional requirements for crossbred lambs with estimated weight gains of 200 g/day. Samples of the ingredients and formulated diets were collected and examined to analyze of their chemical composition (Tables 1 and 2).

Samples (triplicate) were pre-dried at 55 °C for 72 h, ground with a Willey mill (Tecnal, Piracicaba City, São Paulo State, Brazil) with a 1-mm sieve, stored in airtight plastic containers (ASS, Ribeirão Preto City, São Paulo State, Brazil), and sealed properly until use in laboratory analyses of the levels of DM (method 967.03 – AOAC, 1990), ash (method 942.05 – AOAC, 1990), CP (method 981.10 – AOAC, 1990), and EE (method 920.29 – AOAC, 1990). The neutral detergent fiber (NDF) content was determined as described by Van Soest et al. (1991) and expressed as inclusive of residual ash, and heat-stable alpha-amylase was not used. Acid detergent fiber (ADF) contents were determined as described by Robertson and Van Soest (1981). Acid detergent lignin (ADL) contents were determined using the AOAC method 973.18 (2002) in which the ADF residue is treated with 72% sulfuric acid. To this end, the neutral detergent boiling residue was incinerated in an oven at 600 °C for 4 h, and a correction for protein content was applied by subtracting the neutral detergent insoluble N content.

The non-fibrous carbohydrate (NFC) content of the ingredients was determined as described by Mertens (1997) and was calculated using the following equation: $NFC = 100–NDF_{ap} - CP - EE - ash$, where NDF_{ap} is neutral detergent fiber corrected for ash and protein. The neutral detergent insoluble N and acid detergent insoluble N values were obtained following the recommendations of Licitra et al. (1996) (Table 1).

To determine the total phenolic content, the conventional Folin-Ciocalteu spectrophotometric method developed by Singleton and Rossi (1965) was used. Aliquots (100 μL) of extracts were added to 750 μL of distilled water, 500 μL of Folin-Ciocalteu reagent and

Table 1
Ingredients and chemical composition of experimental diets.

Item	Tamarind residue (g/kg as fed)			
	0.00	100	200	300
Ingredient proportions (g/kg DM)				
Cassava silage with added tamarind residue	500	500	500	500
Ground corn	468	407	396	370
Soybean meal	32.0	93.0	105	130
Chemical composition (g/kg DM)				
Dry matter (g/kg as fed)	266	303	375	425
Crude protein	151	152	152	153
Ether extract	38.8	31.2	28.6	27.3
Ash	81.6	84.1	76.0	73.8
Neutral detergent fiber[a]	284	301	314	323
Acid detergent fiber	169	198	207	218
Neutral detergent insoluble protein (g/kg of CP)	132	131	132	128
Acid detergent insoluble protein (g/kg of CP)	121	120	121	117
Acid detergent lignin	26.1	53.8	71.4	84.4
Non-fibrous carbohydrates	445	431	430	423
Total tannins (GAE mg/g DM)[b]	12.9	19.2	23.9	27.3

[a] corrected for ash and protein content.
[b] GAE = gallic acid equivalent.

Table 2
Chemical composition, pH, effluent loss, gas loss, dry matter recovery and ammonia nitrogen of the cassava silage with the tamarind residue additive after opening the silo after 56 days.

Item	Tamarind residue (g/kg as fed)				SEM[a]	*P*- value	
	0.00	100	200	300		Linear	Quadratic
Dry matter (g/kg as silage)	280	318	395	448	16.0	< 0.001	< 0.001
Crude protein (g/kg DM)	208	158	149	128	7.01	< 0.001	< 0.001
Ether extract (g/kg DM)	34.4	21.3	16.3	14.6	2.04	< 0.001	< 0.001
Ash (g/kg DM)	100	98.0	79.4	71.5	3.29	0.018	0.044
NDF_{ap}[b] (g/kg DM)	464	503	530	550	8.45	< 0.001	< 0.001
Acid detergent fiber (g/kg DM)	310	363	380	400	8.88	< 0.001	< 0.001
Acid detergent lignin (g/kg DM)	39.8	97.9	132	162	10.5	< 0.001	0.029
Non-fibrous carbohydrates (g/kg DM)	194	219	225	235	5.21	< 0.001	0.058
Total tannins (GAE[c]/g DM)	26.0	38.3	47.8	54.5	1.54	< 0.001	< 0.001
pH	4.04	3.89	3.85	3.86	0.02	< 0.001	< 0.001
Effluent loss (kg/t FMs)[d]	0.22	0.23	0.22	0.23	0.02	0.29	0.34
Gas losses (g/kg DM)	12.3	15.6	21.4	21.1	15.0	0.059	0.15
Dry matter recovery (g/kg of feed)	990	991	991	991	30.0	0.30	0.34
Ammonia nitrogen (mg/g N)	41.2	34.3	36.2	29.3	14.0	< 0.001	0.99

[a] Standard error of the mean.
[b] Neutral detergent fiber corrected for ash and protein content.
[c] GAE = gallic acid equivalent.
[d] FMs = forage mass in the silage.

1000 μL of 350 g/kg sodium carbonate (Na_2CO_3). The mixture was shaken vigorously after being diluted to 10 mL with distilled water. The mixture was incubated for 30 min at room temperature and read at 725 nm using a GENESYS 10S UV–vis spectrophotometer. Distilled water was used as the blank. Gallic acid standard solutions were prepared (Tamilselvi et al., 2012), and total tannin contents were expressed as GAE mg/g DM (GAE = gallic acid equivalent), as calculated from a standard curve prepared using 0–100 mg/GA (Table 1).

To determine the digestibility of the nutrients in the middle of the trial period, days 37–46, 24 animals were housed in metabolic cages (6 animals from each treatment at this stage). The animals went through an adjustment period of 7 days, and from days 44–46, the orts, provided diet, feces and urine were collected daily. Collected samples were then frozen for further analysis. To collect total feces, appropriate bags were attached to the animals, and samples were collected twice a day, at 08:00 and 15:00 h. They were then weighed and homogenized, and approximately 100 g/kg of the total sample volume was retained for subsequent laboratory analysis. Urine was collected using a bucket collector in the metabolic cage, which contained 0.036 N H_2SO_4 in a proportion equal to 100 g/kg of the total volume of urine from the previous day.

The DM digestibility coefficients (DMDCs) for CP, ether extract (EE), NDF, and NFCs were calculated using the equation: DMDC = [(kg of ingested fraction − kg of excreted fraction)/(kg of fraction ingested)] × 100. The N contents of the triplicate

samples of the provided diet, feces and urine were determined according to the methodology described by the AOAC (1990). The retention of N (N retained, g/d) was determined using the following formula: N retained (g/g) = N intake (g/d) − [N feces excretion (g/d) + N urine excretion (g/d)].

The nutrient intake was determined by subtracting the amount of each nutrient contained in the refusals from the total of each nutrient in the feed offered. The animals were individually weighed at the beginning of the experiment and again every 21 days to determine the performance of lambs and their average daily gain (ADG). The weight measurements were performed in the morning before the first daily feeding and after a continuous fasting period of approximately 16 h.

Feed conversion (feed/gain) was determined using the average dry-weight intake of lambs fed the different diets divided by their ADG, which was calculated based on the difference in the initial and final body weights of animals divided by the number of days in the trial period. Values were expressed as g/g.

At the end of the experiment, the animals were fasted for 16 h and then weighed to determine their slaughter weight. Later, the animals were stunned with a pneumatic pistol and bled. After slaughter, the gastrointestinal tract contents (GC) were removed to determine the empty body weight. After skinning, gutting, and the removal of the head, feet and genitals, the hot carcass weight (HCW, kg) was obtained. The carcasses were cooled in cold storage for 24 h at 4 °C and then weighed to determine the cold carcass weight (CCW, kg). Using these data, the hot carcass yield (HCY) and cold carcass yield (CCY) were calculated based on the carcass weight relative to the body weight at slaughter (g/kg).

Cooling losses [CL (g/kg) = (HCW − CCW)/HCW × 100] and true yield [TY (g/kg) = (HCW/GC) × 100] were determined as described by Cartaxo et al. (2009). Perirenal fat, which was the fat that covered the kidneys, was removed and weighed individually.

In the section between the 13th rib and the 1 st lumbar vertebra, measurements were made to calculate the *Longissimus dorsi* area (LDA). Geometric measurements were made using digital calipers to measure the width of the eye loin (A), which was the maximum width of the muscle from the medial to the lateral edge of the thoracic and lumbar *Longissimus*, and the depth (B), which was the maximum distance perpendicular to the width at the location adjacent to the lateral edge of the vertebrae. Then, the LDA was calculated based on the following equation: (A/2 × B/2) × π, using π = 3.14.

2.4. Statistical analysis

The gas production *in vitro* technique trial was carried out as a completely randomized design in a factorial arrangement 4 × 17 (tamarind residue levels × incubations times) with five replicates (mini-silos) per treatment, according to the mathematical model $Yijk = \mu + \beta i + Rj(\beta i) + \tau j + (\beta^*\tau)ij + Eijk$, where Yij = value observed in the treatment and replicate j; μ = general mean; βi = effect of treatment i = 0.00, 100, 200 or 300 g/kg tamarind residue (as fed); Rj(βi) = random effect based on replication within the treatment; τj = incubation time effect, j = 2, 4, 6, 8, 9, 11, 12, 14, 17, 20, 24, 28, 34, 48, 72, 96 and 120 h; (β*τ)ij = effect of the interaction between treatment and incubation time; Eijk = random error.

The silage quality, digestibility and performance trials followed a completely randomized design with 5, 6 and 10 replicates (animals) per treatment, respectively, according to the mathematical model: $Yij = \mu + \alpha i + Eij$, where Yij = value observed in the treatment and replicate j; μ = general mean; i = effect of treatment i = 0.00, 100, 200 or 300 g/kg tamarind residue (as fed); j = replicate number; and Eij = random error.

Statistical analyses were performed using PROC GLM in SAS version 9.0 (SAS, 2013). Analysis of variance was carried out, and an orthogonal partition of the sum of the square of treatments into linear and quadratic degree effects was obtained. The regression equation was adjusted when significance was $P \leq 0.05$ using PROC REG of SAS version 9.0 (SAS, 2013). Trends were discussed at $P \leq 0.05$ to $P \leq 0.10$.

3. Results

3.1. Changes in silages due to the addition of tamarind residue

The addition of tamarind caused a linear and quadratic increase in the DM ($P < 0.001$) NDF_{ap} ($P < 0.001$), ADF ($P < 0.001$), NFC ($P < 0.001$), lignin ($P < 0.001$) and total tannin ($P < 0.001$) contents of the cassava silage (Table 2). In addition, there was a linear and quadratic reduction in the CP ($P < 0.001$), EE ($P < 0.001$) and ash (linear, $P = 0.018$; quadratic, $P = 0.044$) contents of cassava silages produced with the addition of tamarind residue. Linear and quadratic effects were not observed on DM recovery rate and effluent loss in the cassava silages with tamarind residue added.

There were linear and quadratic reductions in silage pH ($P < 0.001$) and ammonia N content (mg/g N total) ($P < 0.001$) with the addition of tamarind residue (Table 2). However, there was trend for a linear increase in gas loss ($P = 0.059$) from the cassava silages with tamarind residue added.

3.2. Results of trials using the in vitro gas production technique

There was a linear and quadratic increase (mL/g DM) in the maximum potential gas production from total carbohydrates (vt) ($P < 0.001$). However, there were also linear and quadratic decreases (mL/g DM) in the gas production rate from NFCs (m_1) ($P < 0.001$), total gas production rate (mt) ($P < 0.001$), hours in the lag phase ($P < 0.001$) and the in vitro dry matter digestibility kinetics over 48 h (IVDMD) ($P < 0.001$) of the cassava silages with the inclusion of tamarind residue (Table 3). There were no effects of tamarind residue inclusion in cassava silage (mL/g DM) on the gas production rate from NFCs (vf_1) or gas production rates from

Table 3
Gas production and in vitro dry matter degradability kinetics of ruminal fermentation of cassava silage with tamarind residue added at different levels.

Variables	Tamarind residue (g/kg as fed)				SEM[a]	*P*- value	
	0.00	100	200	300		Linear	Quadratic
vf_1 (mL/g DM)[b]	47.9	49.5	55.4	49.3	1.08	0.32	0.11
vf_2 (mL/g DM)[c]	26.0	24.6	26.1	31.4	1.11	0.075	0.055
m_1 (mL/g DM)[d]	0.12	0.11	0.10	0.07	0.01	< 0.001	< 0.001
m_2 (mL/g DM)[e]	0.02	0.02	0.02	0.02	0.01	0.22	0.45
vt (mL/g DM)[f]	73.9	74.1	81.5	80.7	1.03	< 0.001	< 0.001
mt (mL/g DM)[g]	0.14	0.12	0.12	0.09	0.01	< 0.001	< 0.001
Lag phase (h)	9.15	8.27	7.72	6.43	0.97	< 0.001	< 0.001
IVDMD (g/kg)[h]	781	684	679	624	1.50	< 0.001	< 0.001

[b]Maximum potential gas production from non-fibrous carbohydrates.
[a] Standard error of the mean.
[c] Maximum potential gas production from fibrous carbohydrates.
[d] Gas production rates from non-fibrous carbohydrates.
[e] Gas production rates from fibrous carbohydrates.
[f] Maximum potential gas production from total carbohydrates.
[g] Total gas production rate.
[h] *In vitro* dry matter digestibility kinetics over 48 h.

fibrous carbohydrates (FCs) (m_2). There was a linear increase in gas production of 27.0 mL/g DM of the cassava silage with 300 g/kg tamarind residue added.

3.3. Animal performance

The inclusion of tamarind residue in the cassava silage had no effect on the intake (g/d) of DM, CP, NFCs or total digestible nutrients (TDNs). However, there was a linear increase in NDF intake ($P = 0.042$) and a linear reduction in EE ($P = 0.038$) intake by lambs fed cassava silage with added tamarind residue. There were no quadratic effects on NDF or EE intake, but there were linear increases in the DM intake (DMI; g/kg BW) ($P < 0.001$) and NDF_{ap} ($P < 0.001$) in lambs fed cassava silage with tamarind residue added. In addition, there were quadratic effects on the DMI and NDF intake (g/kg BW) of the animals.

There were positive linear and quadratic effects on the DMDC ($P < 0.001$), CP ($P < 0.001$) and reduction of NDF_{ap} ($P < 0.001$) (Table 4) and positive quadratic effects on NFC ($P = 0.066$) and EE ($P = 0.018$) digestibility due to the inclusion of

Table 4
Feed intake and the coefficients of digestibility and nitrogen balance of lambs fed cassava silage with tamarind residue added at different levels.

Item	Tamarind residue (g/kg as fed)				SEM[a]	*P*- value	
	0.00	100	200	300		Linear	Quadratic
Daily feed intake (g/day)							
Dry matter	1011	1098	1115	1144	96.9	0.32	0.75
Crude protein	152	167	170	175	13.8	0.28	0.73
Neutral detergent fiber	287	331	349	370	28.6	0.042	0.68
Non-fibrous carbohydrates	450	474	479	484	38.9	0.54	0.81
Ether extract	39.2	34.3	32.0	31.1	2.66	0.038	0.45
Total digestible nutrients	745	806	818	766	63.2	0.79	0.38
Daily feed intake (g/kg BW)							
Dry matter	32.8	36.5	35.2	38.9	0.50	< 0.001	0.97
Neutral detergent fiber	9.70	11.5	11.6	13.2	0. 20	< 0.001	0.76
Digestibility coefficient							
Dry matter	0.74	0.75	0.75	0.67	0.015	< 0.001	< 0.001
Crude protein	0.52	0.61	0.54	0.40	0.019	< 0.001	< 0.001
Neutral detergent fiber	0.59	0.56	0.60	0.49	0.022	< 0.001	0.10
Non-fibrous carbohydrates	0.92	0.94	0.94	0.91	0.014	0.55	0.066
Ether extract	0.68	0.74	0.70	0.64	0.024	0.19	0.018
Nitrogen Balance (g/d)							
N Intake	26.1	26.9	28.1	28.0	1.06	0.40	0.78
Fecal N excretion	9.54	7.87	10.0	14.1	0.79	< 0.001	0.029
Urinary N excretion	9.39	10.5	10.2	8.61	0.35	0.31	0.018
N Retained	7.13	8.59	7.82	5.27	0.35	< 0.001	< 0.001

[a] Standard error of the mean.

Table 5
Performance and carcass characteristics of castrated lambs fed cassava silage with tamarind residue added at different levels.

Variables	Tamarind residue (g/kg as fed)				SEM[a]	*P*- value	
	0.00	100	200	300		Linear	Quadratic
Initial body weight (kg)	21.2	22.6	21.5	23.2	–	–	–
Final body weight (kg)	29.7	30.7	31.3	30.9	0.61	0.21	0.33
Average daily gain (g/d)	105	119	127	122	4.90	0.21	0.33
Feed conversion (g/g)[b]	9.57	9.80	9.16	10.2	0.33	0.60	0.44
Hot carcass weight (kg)	15.6	16.1	17.2	16.9	0.38	< 0.001	0.28
Hot carcass yield (g/kg)	524	526	549	546	3.60	< 0.001	0.65
Cold carcass weight (kg)	15.1	15.6	16.6	16.4	0.37	< 0.001	0.37
Cold carcass yield (g/kg)	508	509	530	530	3.40	< 0.001	0.94
True yield (g/kg)	601	601	621	609	3.20	0.12	0.45
Cooking weight losses (g/kg)	30.2	31.9	35.0	29.1	3.20	1.00	0.25
Perirenal fat (kg)	1.03	1.13	1.07	1.29	0.06	0.16	0.90
Longissimus dorsi area (cm)	9.43	10.6	10.9	11.4	0.26	< 0.001	0.20

[a] Standard error of the mean.
[b] Feed/gain ratio.

tamarind residue in cassava silage.

The addition of tamarind residue in cassava silage resulted in a linear and quadratic increase in fecal N excretion (linear, $P < 0.001$; quadratic, $P = 0.029$) and a quadratic increase in urinary N excretion ($P = 0.018$) in lambs. For retained N ($P < 0.001$) in lambs, the results were the opposite: a linear and quadratic reduction was observed with the inclusion of tamarind residue in cassava silage. There was no linear or quadratic effect of tamarind residue inclusion in cassava silage on N intake.

There were no linear or quadratic effects in relation to the levels of tamarind residue included in the cassava silage on final body weight, average daily weight gain, feed conversion (feed/gain), true carcass yield, cooking loss and perirenal fat (Table 5). However, HCW ($P < 0.001$), HCY ($P < 0.001$), CCW ($P < 0.001$), CCY ($P < 0.001$) and LDA ($P < 0.001$) all showed a linear increase with increased inclusion levels of tamarind residue in the cassava silage, whereas there were no quadratic effects in relation to the different diets.

4. Discussion

4.1. Silage

The absorptive effect of tamarind residue was demonstrated by the greater levels of DM in the cassava silages. The ideal level of DM in silage described by McDonald et al. (1991) is approximately 280–350 g/kg. A value in this range was observed with the inclusion of tamarind residue at 100 g/kg (DM of silage = 318 g/kg), and the DM content was even greater with the addition of tamarind residue at 300 g/kg to the silage (DM of silage = 448 g/kg). Therefore, tamarind residue functioned as an absorptive additive by adjusting the DM content of the cassava silage to levels that enabled proper fermentation to occur. This was beneficial because it prevented the high humidity needed for the development of bacteria from the genus *Clostridium* responsible for butyric fermentation, which is the main cause of losses in silage (McDonald et al., 1991).

However, the inclusion of tamarind residue at 300 g/kg cassava silage had some negative effects on the silage, such as a reduction in the CP (80 g/kg), EE (20 g/kg) and ash (30 g/kg) contents and increases in the NDF_{ap} (90 g/kg), ADF (90 g/kg) and lignin (125 g/kg) contents, as well as in the tannin (28 GAE/g DM) content; tannins are phenolic, non-nutritional components in the diets of ruminants that limit the ruminal degradation of the fibrous fraction of feed and impair CP digestibility.

Furthermore, silage with a high level of CP is beneficial because it allows for increased proteolysis, which results in a buffering effect that hinders the reduction of the pH to levels that are optimum for fermentation (Napasirth et al., 2015). In addition, the ammonia content of the silage was also reduced by 12 mg/g N with the addition of tamarind residue and ranged from 41.2 to 29.3 mg/g N. The ammonia N level reflects the extent of proteolysis during fermentation in the silo. However, the reduction of ammonia indicates that protein preservation during acidogenesis, which is a relevant outcome because cassava has a high protein content (205 g/kg). Van Soest (1994) reported that values of ammonia N below 100 mg/g of total N are considered adequate for fermentation.

The increase in the NDF_{ap} and ADF values is very important, as the levels of these components indicate the degree of degradability of forage, which according to Van Soest (1994) can limit the DMI of animals via physical mechanisms. Forage with NDF levels greater than 600 g/kg is considered to be of low quality. The present results show that despite the increase in residue levels, NDF_{ap} from tamarind residue (550 g/kg) remained below the maximum limits recommended for high-quality silage.

The inclusion of tamarind residue resulted in pH values ranging from 4.04 to 3.85, which are acceptable for providing aerobic stability in the silage because suitable pH values range from 3.8 to 4.2 (Massafera et al., 2015). This reduction in pH was also promoted by the increase in NFC (180 g/kg) content, which put this parameter at the minimum threshold for changing the type of fermentation. This reduction also promoted increased effluent and gas losses.

4.2. Trial using the in vitro gas production technique

The vt increased, probably due to the carbohydrate content present in cassava waste. With the addition of tamarind residue, there was an increase in the fermentable substrate in the cassava silage. This is because the fermentation of FCs, such as cellulose and hemicellulose, produces a greater proportion of acetate, which results in a greater proportion of substrates that can be used by methanogenic bacteria (Medeiros et al., 2015; Morais et al., 2015). This occurs because the rumen microbial degradation of carbohydrates converts them primarily to glucose-1-P, which is subsequently oxidized to pyruvic acid by the Embden-Meyerhof cycle and thereafter to acetate and propionate by the action of the pyruvate lyase enzyme (Van Soest, 1994).

There was a reduction of 0.14 mL of gas produced from NFCs "m_1" (ml/g DM) with the inclusion of 100 g/kg tamarind residue in cassava silage. The reduction in the NFC gas production rate occurred because there was a marked increase in fiber content and ADL with the inclusion of tamarind residue. The greatest gas production rate occurred at the initial stage of fermentation when the high concentration of NFC resulted in an increased fermentation rate. Over time, these components became scarce, and the remaining energy source, NDF, was fermented at a slower rate (Napasirth et al., 2015).

There was a reduction of mt (ml/g DM) of 0.16 mL with the inclusion of 100 g/kg tamarind residue in the cassava silage. The decrease can be explained by the increase in tannins in the residue complexing with proteins, which may reduce the availability of nutrients for fermentation and consequently depress the rate of carbohydrate degradation (Vázquez et al., 2016; Wang et al., 2016). This fact also explains the reduction that occurred in DMD *in vitro*.

Phase latency or lag time is the time between the start of incubation and the start of microbial action in an incubated sample and is related to the degradation of fibers (Silva et al., 2015b). This parameter showed a linear effect, which can be attributed to the NFC content of the residue (268 g/kg). According to Mertens (2002), NFC, which is represented by water-soluble sugars (mono and disaccharides), starch and pectin, are fermented quickly and are completely digestible in the gastrointestinal tract, which results in shorter colony growth times due to the rapid degradation and fermentation of this soluble fraction.

4.3. Animal performance

The increase of 200 g/kg in the DMI of the animals receiving silage with greater levels of included tamarind residue was due to an increase of 400 g/kg in the DM content of the silage. This shows that the increases in the fiber and the phenolic (lignin and tannin) contents caused by the inclusion of the tamarind residue in the silage did not cause a physical decrease in DMI (Bezerra et al., 2015).

The inclusion of tamarind residue in cassava silage, up to 300 g/kg of DM, increased DMI to 38.9 g/kg of BW, which is above the intake recommended (approximately 30 g/kg BW) by the NRC (2007). Ben Salem et al. (2000a) emphasized that tannins interfere with voluntary DMI, which suggests that the effects found in this study may have been minimized by the use of solid waste (tamarind residue) in cassava silage, which according to McSweeney et al. (2001) reduces the effects of tannins in the diet because of the acidic and anaerobic conditions of the silage.

The inclusion of tamarind residue in cassava silage at levels up to 200 g/kg of the total DM in the diets of lambs contributed to an increased efficiency in DM digestibility. However, greater levels tended to depress the digestibility of this fraction, possibly due to the increases in tannin and lignin concentrations, which can have toxic effects on ruminal microorganisms (Geron et al., 2015). According to Orlandi et al. (2015), increasing dietary levels of tannin significantly decreased the apparent nutrient digestibility. Wang et al. (2016) observed that the digestibility of DM, CP, and EE was 570 g/kg, 870 g/kg and 860 g/kg, respectively, when tamarind residue was included in sheep diets, and a higher level (832 g/kg) of NFC digestibility was observed. However, according to Hristov et al. (2013), beneficial results can be achieved when tannins are added to the diets of ruminants at appropriate levels that favor and promote increased efficiency in the ruminal digestion process.

The inhibitory properties of tannins depend on the total concentration of proteins (Patra and Saxena, 2011). The mechanism postulated to tannin content is that tannins complex proteins at the pH of the rumen and protect them from microbial enzymes. Thus, there is an increase in the efficiency of microbial protein synthesis and a decrease in the feed protein degradability (Van Soest, 1994), that is beneficial for ruminants as they increase the supply of non-ammonia N to the lower intestine (Makkar, 2003). The duodenal flux of all amino acids is evidently improved by tannins (Theodoridou et al., 2010). These effects lead to protein-sparing effects in ruminants, reductions in methane production (Hassanat and Benchaar, 2013) and N excretion to the environment (Lapierre et al., 2012), which thereby reduce the emission of environmental pollutants. In addition, tannins can form complexes with indigestible cell-wall carbohydrates and can bind to bacterial enzymes, by which they reduce the activities of ruminal microorganisms such as cellulolytic bacteria (Geron et al., 2015; Gonçalves et al., 2015).

TDN intake ranged from 745 to 818 g/day at levels of 0–300 g/kg tamarind residue included in the cassava silage, respectively. Microbial protein production and flow into the abomasum is related to the use of TDNs. According to the NRC (1996), for each 100 g of TDN intake, 13 g of microbial proteins is produced. Thus, it was estimated that for diets containing 0.00, 100, 200 and 300 g/kg of tamarind residue, 97.0, 115, and 112, and 109 g of microbial CP, respectively, would be produced per day in the duodenum. Based on this information, it is expected that the addition of tamarind residue in cassava silage contributes to muscle growth and consequently to better animal performance.

The lack of effect on body weight at slaughter can be explained by the fact that animals presented similar average daily weight gains and did not show an effect of diet on feed conversion. In another analysis, it was possible to infer that there was a more efficient use of the retained N because, even with the lower N retention levels in lamb fed the diets with the highest levels of tamarind residue, the animals gained similar amounts of weight at all inclusion levels (Agy et al., 2012). Moderate concentrations (20 and 40 g/kg in DM basis) of condensed tannins may benefit the nutrition and health of small ruminants (Ben Salem et al., 2000b). This is because at

these levels, tannins bind to certain proteins and protect them from excessive ruminal degradation (Hassanat and Benchaar, 2013). These tannin-bound proteins pass through the rumen and are released into the duodenum, the portion of the gastrointestinal tract in which the absorption of amino acids occurs more intensely, which results in better utilization of dietary protein (Theodoridou et al., 2010). Diets with greater amounts of tannins, due to the inclusion of tamarind residue in the ensiling process, influence the parameters of ruminal kinetics, with a reduction in gas production rates that results in lower energy loss from animals in the form of gases lost to the environment. This indicates a greater increase in weight in the form of muscle and body fat and thus affects carcass yield (Wang et al., 2016). Note that the HCY and CCY were obtained through mathematical calculations involving the live weight at slaughter after fasting, empty body weight, HCW and CCW. Depending on dietary passage rates, the HCY and CCY vary as a function of gastrointestinal tract filling (Oliveira et al., 2015b).

5. Conclusions

The addition of 300 g/kg tamarind residue in the ensiling of cassava improves the quality of cassava silage because it increases the DM content and reduces the production of gases. Therefore, the inclusion of tamarind residue in the ensiling of cassava in lamb diets is recommended because including this silage as roughage (500 g/kg of feed) contributes to increased DMI and DM digestibility, N retention, HCY, CCY, and LDA.

Conflict of interest

The authors have no conflicts of interest to declare.

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