DOI: 10.5433/1679-0359.2023v44n2p929

# Mixed sorghum and forage cactus silage: composition, digestibility, fermentation, and losses

# Silagem mista de sorgo e palma forrageira: composição, digestibilidade, fermentação e perdas

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## Highlights \_

Cactus affects the composition, digestibility, and fermentation of sorghum silage. ≥ 20% forage cactus levels may reduce sorghum silage fermentability. 90:10 ratio is recommended for sorghum:cactus mixed silage production.

## Abstract

The production of mixed silages can be an essential strategy for storing and processing feed for ruminants in arid and semi-arid regions. This study aimed to evaluate the effect of inclusion levels of forage cactus in sorghum silage on chemical-bromatological composition, losses, in vitro digestibility, and fermentative profile. Sorghum silages were produced by adding 0, 10, 20, 30, and 40% forage cactus based on natural matter. Experimental silos were filled with the mixtures and hermetically sealed. The silos were opened after 34 days of fermentation, and the samples were analyzed for composition, digestibility, fermentative profile, and losses in silage. The inclusion of forage cactus presented a negative quadratic influence (P < 0.05) on the contents of dry matter (DM), ether extract, neutral detergent fiber, acid detergent fiber, and cellulose of sorghum silage. The DM concentration decreased from 35.83 to 25.43% for the control treatment (0%) to the 20% treatment, followed by stabilization at values close to 26 ± 1% in subsequent levels. Digestibility *in vitro* and the total digestible nutrients of the silages increased linearly (P < 0.05) with the inclusion of forage cactus. However, there was a linear increase (P < 0.05) of pH, ammonia nitrogen,

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acetic acid, and butyric acid with the inclusion of forage cactus, reaching values indicative of the limitation of the fermentation process from the level of 20% inclusion. Including forage cactus in sorghum silage did not affect (P > 0.05) losses by gases, effluents, and DM recovery from silage. Adding up to 10% of forage cactus can benefit chemical-bromatological characteristics, digestibility, and fermentation of sorghum silage.

Key words: Ensiling. Nopalea cochenillifera L. Organic acids. Silage moisture.

#### Resumo .

A produção de silagens mistas pode ser importante estratégia para armazenamento e beneficiamento dos alimentos para ruminantes em regiões áridas e semiáridas. Objetivou-se avaliar efeito de níveis de inclusão de palma forrageira na ensilagem do sorgo sobre composição químico-bromatológica, perdas, digestibilidade in vitro e perfil fermentativo. Foram produzidas silagens de sorgo com adição de 0, 10, 20, 30 e 40% de palma forrageira com base na matéria natural. Utilizou-se silos experimentais preenchidos com as misturas e fechados hermeticamente. Após 34 dias de fermentação os silos foram abertos e as amostras analisada quanto a composição, digestibilidade, perfil fermentativo e perdas na ensilagem. A inclusão de palma forrageira influenciou de forma quadrática negativa (P < 0,05) os teores de matéria seca (MS), extrato etéreo, fibra em detergente neutro, fibra em detergente ácido e celulose da silagem de sorgo. The DM concentration decreased from 35.83 to 25.43% for the control treatment (0%) to the 20% treatment, followed by stabilization at values close to 26 ± 1% in subsequent levels. A digestibilidade in vitro e os nutrientes digestíveis totais das silagens aumentaram linearmente (P < 0.05) com a inclusão de palma forrageira. Contudo, houve incremento linear (P < 0,05) do pH, nitrogênio amoniacal, ácido acético e ácido butírico com a inclusão de palma forrageira, atingido valores indicativos de limitação do processo fermentativo a partir do nível de 20% de inclusão. A inclusão de palma forrageira na ensilagem do sorgo não influenciou (P > 0,05) as perdas por gases, efluentes, e recuperação de MS da silagem. A adição até 10% de palma forrageira pode ser recomendada para beneficiar características químico-bromatológica, digestibilidade e fermentação da silagem de sorgo.

Palavras-chave: Ácidos orgânicos. Ensilagem. Nopalea cochenillifera L. Umidade na silagem.

#### Introduction \_\_\_\_

The rainfall instability of arid and semi-arid zones is the primary cause of the variation in pasture phytomass observed in these regions. Under these conditions, using voluminous conservation strategies, such as silage, should increase the food security of the herds and improve zootechnical indices throughout the year. The temporal and spatial variations of the rainfall regime also make it challenging to grow forage for silage. Thus, even adapted forages, such as sorghum, may present variations in composition and productivity level (Moura et al., 2016). The production of mixed sorghum silages with other droughttolerant species, such as forage cactus, can be an alternative to circumvent the vulnerability of production systems in arid and semi-arid regions.

The forage cactus is a cactus well adapted to arid zones. It presents 11.78 ± 1.79% dry matter, 4.32 ± 0.93% crude protein, 53.74 ± 2.18% non-fibrous carbohydrates (NFC), 29.08 ± 1.57% in neutral detergent fiber (NDF), and about 63.43% of total digestible nutrients (Valadares et al., 2018). The high NFC/NDF ratio represents a good condition for the association of forage cactus with fiberrich feeds (Voltolini et al., 2016). However, it is necessary to determine the inclusion levels of cactus in association with fibrous feeds, especially for silage production, due to its high moisture content. Borges et al. (2020) reported relevant effects when studying cactus and elephant grass mixed silages, with increases in the contents of residual soluble carbohydrates and a decrease in pH with the inclusions of 20, 40, and 60% of cactus.

Therefore, this study aimed to evaluate the effects of the inclusion of forage cactus in forage sorghum silage on the chemical-bromatological composition, *in vitro* digestibility, losses, and fermentative profile of silages.

The experimental design was completely randomized, with four replicates per treatment. The treatments consisted of levels of addition of forage cactus (0%, 10%, 20%, 30%, and 40%) in sorghum silage based on natural matter. Sorghum bicolor cv. BR 700 was used in the maturation stage of milky/ pasty grain, with an approximate age of 100 days, grown on a rural property in Governador Dix-Sept Rosado, RN, Brazil. Mature cladodes of clone IPA Sertânia forage cactus (Nopalea cochenillifera L. Salm-Dyck) were obtained from the same property. The region's climate is classified as BSH low latitude and altitude by the Köppen criteria.

All feed was finely ground into particles 1-2 cm thick and mixed in the proportions described earlier. Aliquots of the mixtures were collected to determine the dry matter (DM) before silage. Subsequently, the mixtures were ensiled under a density of 500 kg m<sup>-3</sup>, and the silos were hermetically sealed. Experimental PVC silos 50 cm in length and 10 cm in diameter with a capacity of approximately 4 kg were used. Bunsen valves were installed in the silos for gas elimination. A layer of paper towels was placed at the bottom of each silo for liquid absorption and retention. After 34 days, sufficient period for stabilizing the anaerobic fermentation process, the silos were opened, and homogenized samples were collected and subdivided for further analysis.

The subsamples intended for chemical-bromatological analysis were predried in a forced air circulation oven at 65 °C for 48 hours, weighed, and ground in a knife mill with a 1 mm sieve. The contents of DM, crude protein (CP), ether extract (EE), and mineral matter (MM) were determined following the respective methods: 934.01, 976.05, 963.15, and 942.05 of the Association of Official Agricultural Chemists [AOAC] (2019).

The contents of acid detergent fiber (ADF) and lignin (LIG), neutral detergent fiber (NDF) corrected for ash and proteins, and cellulose (CEL) and hemicellulose (HEM) were analyzed and estimated according to AOAC (2019), using methods 973.18 and 2002.04. Soluble carbohydrates (SCHO) were determined according to Bailey (1967).

The pre-dried subsamples were subjected to the *in vitro* dry matter digestibility (IVDMD) testing method proposed by Tilley and Terry (1963) using a Daisy II incubator (ANKOM<sup>®</sup> Technology), following adaptations proposed by Holden (1999). We used ruminal inoculum collected through a cannula in an adult cow (650 kg body weight) fed daily with forage sorghum silage and concentrated feed. This procedure was approved by the Ethics Committee on Animal Use of the Universidade Federal Rural do Semiárido, Protocol nº 23091.010626/2019-23.

Non-fibrous carbohydrates (NFC) were estimated according to Mertens (1997). Total digestible nutrients (TDN), digestible energy (ED), and metabolizable energy (ME) were estimated as proposed by Detmann et al. (2016).

The losses during processing were estimated as described by Jobim et al. (2007). The losses by gas (LG) were estimated by the difference between the initial and final weight of the closed silos. The losses by effluents (LE) were estimated by the difference in weight of the paper towels before and after the ensilage process. DM recovery (DMR) was calculated by the following relationship: DMR = 100 - (% DM at silo closure - % DM at silo opening).

The fresh subsamples were manually pressed to extract residual liquid from the silage. The pH and ammonia nitrogen (N-NH<sub>3</sub>), as recommended by Mizubuti et al. (2009), and the concentrations of organic acids, according to Erwin et al. (1961), were analyzed in the extracted liquid. The acids identified were acetic acid (AA), propionic acid (PA), butyric acid (BA), and lactic acid (LA).

Orthogonal polynomial contrasts were applied to verify the linear and quadratic effects of the increase of the inclusion of forage cactus in sorghum silage on the variables studied. The SAS PROC GLM procedure was used, and linear and quadratic effects were considered significant with P < 0.05.

The inclusion of forage cactus influenced (P < 0.05) the chemicalbromatological composition of sorghum silage (Table 1). The increase in the inclusion of forage cactus generated an increasing linear effect (P < 0.05) on the concentrations of MM, CP, SCHO, TDN, DE, DM, and IVDMD.

The concentrations of DM and EE were adjusted (P < 0.05) to negative guadratic polynomial regression models with increased levels of inclusion of forage cactus in sorghum silage. The inverse was observed in the concentration of NFC, which showed a positive quadratic effect on the increase in forage cactus. The DM concentration reduced by 5.4% in the control treatment (0% cactus inclusion) up to level 20, reducing from 35.83 to 25.43%, followed by stabilization at values close to  $26 \pm 1\%$  in subsequent levels. The NFC content of the silage increased from 31.05% of the control to 41.24% at level 10, followed by a gradual reduction until reaching 39.80% at level 40.

The DM content responses are essential factors to consider in evaluating silage since it represents the set of potentially digestible feed nutrients. In addition, concentrations of less than 25% DM in the silage material of grasses represent a risk of *Clostridium* bacteria (Kung et al., 2018). The control of the activity of undesirable microorganisms in the silage depends mainly on the reduction of pH to levels below 4.5 and the increase in osmotic pressure (DM greater than 25%), which should inhibit the action of Clostridium and Enterobacteria (Muck et al., 2018).



There was a negative quadratic effect (P < 0.05) of the inclusion of forage cactus on the concentrations of NDF, ADF, and CEL of the silage (Table 1), with minimum points estimated at 43.00, 30.83, and 20.00,

respectively. There was a decreasing linear (P < 0.05) effect of the inclusion of forage cactus on the concentrations of HEM and LIG in sorghum silage (Table 1).

#### Table 1

Effect of forage cactus inclusion levels on dry matter (DM), mineral matter (MM), crude protein (CP), ether extract (EE), neutral detergent fiber (NDF) ash and protein-free, acid detergent fiber (ADF), cellulose (CEL), hemicellulose (HEM), lignin (LIG), soluble carbohydrates (SCHO), non-fiber carbohydrates (NFC), *in vitro* dry matter digestibility (IVDMD), total digestible nutrients (TDN), digestible energy (DE) and metabolizable energy (ME) of sorghum silage

Variable –	Forage cactus inclusion levels (%)						<i>P</i> value	
	0	10	20	30	40	SEM <sup>1</sup>	L <sup>2</sup>	Q³
DM (%) <sup>4</sup>	30.83	29.06	25.43	27.78	25.92	0.47	0.0007	0.0201
MM (% DM)⁵	7.61	9.41	9.67	9.87	10.43	0.22	<.0001	0.0022
CP (% DM) <sup>6</sup>	6.85	7.07	7.44	7.61	7.94	0.09	0.0001	0.9895
EE (% DM) <sup>7</sup>	2.85	1.64	2.17	2.70	2.91	0.12	0.0058	0.0013
NDF (% DM) <sup>8</sup>	51.65	40.64	40.19	39.37	38.93	1.10	<.0001	<.0001
ADF (% DM) <sup>9</sup>	44.46	20.32	20.09	20.04	19.43	2.25	<.0001	<.0001
CEL (% DM)10	33.32	28.24	28.10	28.62	28.58	0.46	<.0001	<.0001
HEM (% DM) <sup>11</sup>	14.77	12.02	11.32	11.10	10.88	0.34	<.0001	<.0001
LIG (% DM) <sup>12</sup>	6.60	5.68	4.46	4.23	4.10	0.23	<.0001	<.0001
SCHO (% DM) <sup>13</sup>	14.75	29.35	29.62	29.70	29.81	1.37	<.0001	<.0001
NFC (% DM) <sup>14</sup>	31.05	41.24	40.53	40.46	39.80	0.88	<.0001	<.0001
IVDMD (%) <sup>15</sup>	53.08	53.93	53.93	55.86	55.49	0.24	0.0067	0.5659
TDN (% DM) <sup>16</sup>	65.96	65.72	67.96	68.74	68.62	0.31	0.0099	0.3849
DE (Mcal/kg DM) <sup>17</sup>	2.82	2.81	2.91	2.95	2.94	0.01	0.0064	0.3683
ME (Mcal/kg DM) <sup>18</sup>	2.36	2.36	2.45	2.48	2.48	0.01	0.0067	0.3756

<sup>1</sup> Standard error of the mean; <sup>2</sup> linear effect; <sup>3</sup> quadratic effect; <sup>4</sup> Y =  $30.85 - 0.28x + 0x^2$  ( $r^2 = 0.74$ ); <sup>5</sup> Y = 7.81 + 0.13x ( $r^2 = 0.78$ ); <sup>6</sup> Y = 6.84 + 0.03x ( $r^2 = 0.95$ ); <sup>7</sup> Y =  $2.62 - 0.07x + 0.002x^2$  ( $r^2 = 0.61$ ); <sup>8</sup> Y =  $50.47 - 0.86x + 0.01x^2$  ( $r^2 = 0.89$ ); <sup>9</sup> Y =  $41.69 - 1.85x + 0.03x^2$  ( $r^2 = 0.86$ ); <sup>10</sup> Y =  $32.72 - 0.40x + 0.01x^2$  ( $r^2 = 0.84$ ); <sup>11</sup> Y = 14.55 - 0.25x ( $r^2 = 0.71$ ); <sup>12</sup> Y = 6.67 - 0.14x ( $r^2 = 0.86$ ); <sup>13</sup> Y = 16.38 + 1.14x ( $r^2 = 0.52$ ); <sup>14</sup> Y =  $32.26 + 0.77x - 0.02x^2$  ( $r^2 = 0.82$ ); <sup>15</sup> Y = 53.03 + 0.08x ( $r^2 = 0.82$ ); <sup>16</sup> Y = 65.55 + 0.12x ( $r^2 = 0.75$ ); <sup>17</sup> Y = 2.80 + 0.01x ( $r^2 = 0.77$ ); <sup>18</sup> Y = 2.35 + 0.01x ( $r^2 = 0.77$ ).

Cactus is an excellent energy source, rich in non-fibrous carbohydrates, and has a high digestibility coefficient (Margues et al., 2017). In general, the responses of the forage cactus increase on the chemical composition of sorghum silage can be considered positive. The results indicate that the inclusion of cactus increased CP, MM, SCHO, NFC, and reduced fibrous constituents of low digestibility, especially ADF and LIG. This relationship improved the energy value (TDN, DE, and ME) and digestibility (IVDMD and IVOMD) of silages. However, the most significant variation in the composition of the silages occurred from the control (0% cactus) to the lowest inclusion level (10% cactus) and in the sequence (from 10 to 20, from 20 to 30, and from 30 to 40%). The linear or quadratic responses were attenuated, with less variation between levels.

The inclusion of forage cactus did not affect (P > 0.05) the concentration of PA, losses by gas (GL) and effluents (EL), and DM recovery (DMR) of silage (Table 2). However, pH and the concentrations of N-NH<sub>a</sub>, AA, and BA showed a linear increase with increasing levels of forage cactus inclusion in sorghum silage (Table 2). Because it is a high-moisture feed containing about 10 to 13% DM (Valadares et al., 2018), the increase in the proportion of forage cactus reduced the DM and increased the BA and pH in sorghum silage. These results indicate that the high moisture conditions reduced the osmotic pressure, attenuating the pH reduction and allowing the activity of butyrate-producing Clostridium in the silages with high cactus inclusion.

#### Table 2

Effect of forage cactus inclusion levels on hydrogen potential (pH), ammonia hydrogen (N-NH3), acetic acid (AA), propionic acid (PA), butyric acid (BA), lactic acid (LA), gas losses (GL), effluent losses (EL), and dry matter recovery (DMR) of sorghum silage

Variable -	Forage cactus inclusion levels (%)					SEM <sup>1</sup>	<i>P</i> value	
	0	10	20	30	40	SEIVI.	L <sup>2</sup>	Q³
pH⁴	3.84	4.12	4.16	4.18	4.21	0.03	<.0001	<.0001
N-NH <sub>3</sub> (% total N) <sup>5</sup>	5.68	6.82	7.48	8.40	8.84	0.27	0.0002	0.1822
AA (% DM) <sup>6</sup>	3.65	4.62	4.63	4.92	4.92	0.11	<.0001	0.0003
PA (% DM) <sup>7</sup>	2.74	1.88	3.54	2.94	3.74	0.18	0.9994	0.4170
BA (% DM) <sup>8</sup>	0.18	0.33	0.58	0.58	0.83	0.06	0.0426	0.8349
LA (% DM) <sup>9</sup>	4.91	4.97	5.06	5.25	5.32	0.04	0.0956	0.4111
GL (% DM) <sup>10</sup>	6.01	3.58	3.35	3.46	3.33	0.50	0.1493	0.2592
EL (kg/t DM)11	1.20	1.87	1.42	1.97	1.95	0.27	0.7025	0.8722
DMR (% DM)12	92.82	95.03	94.77	94.79	95.29	0.50	0.3446	0.5056

<sup>1</sup> Standard error of the mean; <sup>2</sup> linear effect; <sup>3</sup> quadratic effect; <sup>4</sup> Y = 3.86 + 0.02x (r<sup>2</sup> = 0.67); <sup>5</sup> Y = 5.70 + 0.11x (r<sup>2</sup> = 0.88); <sup>6</sup> Y = 3.74 + 0.08x (r<sup>2</sup> = 0.69); <sup>7</sup> Y = 2.51; <sup>8</sup> Y = 0.18 + 0.02x (r<sup>2</sup> = 0.74); <sup>9</sup> Y = 4.90; <sup>10</sup> Y = 5.67; <sup>11</sup> Y = 1.31; <sup>12</sup> Y = 93.25.

In addition to raising the BA content to critical levels in the silage, the reduction of DM content with the inclusion of cactus could cause greater losses in the silage process. However, GL, EL, and DMR were not influenced, and BA was kept below 1%. These results are related to the control of experimental conditions. Additionally, the reduction of pH is essential for the preservation of nutrients present in the silage material. During ensilage, the SCHO are fermented to organic acids. The LA produced by lactic bacteria contributes most to reducing the final pH, which should be 3.5 to 4.2 in grass silages (Kung et al., 2018). The linear increase in pH with the inclusion of cactus oil did not significantly exceed the recommended range. However, the highest levels (20, 30, and 40%) showed critical values (4.16, 4.18, and 4.21, respectively).

Higher final pH values of the silages that received higher levels of cactus are related to a lower rate of pH decrease, which is related to the increases observed in AA and BA. The higher concentration of AA and AB are associated with the prolonged action of Enterobacteria and Heterofermentative lactic acid bacteria, which negatively affect the pH reduction by producing less LA in relation to Homofermentative bacteria (Muck et al., 2018). In addition, the clostridial metabolism from LA to BA due to the increase in silage moisture (Kung et al., 2018) can explain the increase in BA associated with the maintenance of LA concentrations due to the inclusion of cactus in sorghum silage.

The addition of forage cactus affects the chemical-bromatological composition, digestibility, and fermentation of forage sorghum silage without influencing losses during ensilage. However, we recommend the inclusion of up to 10% cactus based on the natural matter to ensure an adequate silage fermentation process. High levels of cactus inclusion would not be recommended in everyday field situations since it would provide DM in a concentration less suitable for the fermentation process.

### Acknowledgments \_\_\_\_\_

To the Universidade Federal Rural do Semi-Árido, Universidade Federal do Acre; and to CAPES for the financial aid to the Strategic Postdoctoral fellow linked to processes 88887.645288/2021-00 and 88887.201281/2023-00.

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