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Research Article

Silage as a Feasible Technique for Calabash Tree (*Crescentia cujete*) Fruit Conservation: Evaluation of Different Mixtures and Alternatives

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This research was aimed at evaluating the efficiency of silage technique with or without additional ingredients for conserving the fruit of the calabash tree (*Crescentia cujete*) as a source of food for cattle in the tropical dry forest. Five treatments were distributed in a completely randomized design, following a 5 × 5 factorial scheme with five treatments and five conservation times (7, 14, 28, 56, and 90 days) in order to understand, not only the effect of treatments but also the effect of time on the assessed fermentation and quality parameters. Hence, experimental treatments were defined as follows: T0, consisting of unground and not ensiled fruits; T1, ensiled ground fruit with no additives; T2, ensiled ground fruit with 1.5% of common salt (NaCl) addition; T3, ensiled mixture of ground fruit and calabash tree foliage in a fresh basis of 30:70 fruit-to-roughage ratio; and T4, ensiled mixture of ground fruit and Angleton (*Dichanthium aristatum*) hay in a fresh basis of 50:50 fruit-to-roughage ratio. The fermentative profile (i.e., pH, buffer capacity, NH_3-N, and organic acid concentrations), nutritional value, and losses (i.e., fresh matter, dry matter (DM), nutrients, and gas) were determined. Not ensiled calabash tree fruit (T0) showed undesirable conservation characteristics, with the highest pH, the highest losses, and the poorest nutritive value after 90 days. Inclusion of forage in silage in treatments T3 and T4 increased the DM and the neutral detergent fiber (NDF) content compared to treatment T1. This dramatically reduced the specific density in treatment T4, which allowed fungus to appear and spoil the silage, resulting in a poor fermentative profile. T1 and T2 showed the lowest total losses, followed by T3. They also showed the highest concentrations of lactic acid and the lowest pH values, despite their high buffering capacity. The hard shells of the fruits were not sufficient to preserve the pulp and its nutritional value for more than 28 days. The addition of common NaCl did not improve the fermentative profile or nutritive value, so it is not necessary. This study has demonstrated the feasibility of silage technique as a method for preserving calabash tree fruit and suggest fruit silage without any additives followed by silage of ground fruit and calabash tree foliage mixture (30:70 fruit-to-roughage ratio) as efficient alternatives for preserving these

feeds to supplement ruminant diets during dry season shortages in tropical farming systems. Further researches are necessary to evaluate the reproducibility and scalability of these results.

Keywords: dry season; fermentation profile; ruminant nutrition; supplementation; tropical tree fruits

Summary

- Ensiling is a necessary and feasible technique for conserving the fruit of the calabash tree without adversely affecting the nutritive value of the final product.
- We failed to prove the locally held belief that the hard shell of the calabash tree fruit effectively preserves its pulp in a 'natural' manner.
- The addition of common salt (NaCl) at 1.5% did not have a significant effect on improving either the conservation or the final nutritive value of the silage.
- The findings indicate that fruit silage is the most reliable ensiling alternative, obviating the need for the incorporation of common NaCl or tropical forages.

1. Introduction

Silvopastoral systems are agroforestry arrangements used as models of sustainable livestock production with high capacity to mitigate the negative effects of climate change [1]. Its benefits include increase of feed supply, carbon sequestration, water retention, and independence on external inputs, which results in more profitable and less environmentally aggressive livestock systems [2–4]. These systems also reduces soil erosion and animal heat stress while increasing biodiversity and biological resilience of the ecosystems [4–7]. The benefits of silvopastoral systems and the emergence of policies which promote payments for environmental services and carbon bonus have led to increase the adoption of these farming systems in recent years [8–11].

Tropical dry Caribbean is characterized by prolonged dry seasons (4–6 months) alternating a rainfall pattern that concentrates more than a half of the annual precipitations in only 2 months, where sometimes, acid, saline, and low fertility soils are part of the livestock farming landscape [12]. Calabash tree (*Crescentia cujete*) is a native plant from the tropical America, recognized by its capacity to thrive in adverse climate conditions [13]. This tree produces fleshy and nutritive fruits (~700 g of weight, yielding 93.4 g/kg DM of CP and 18.1 MJ/kg DM of GE; Table 1) that have traditionally been used by local farmers as a livestock feeding resource, mainly during the dry season for supplying the forage deficit [15]. In addition, fruits and foliage of the calabash tree have been reported as rich in bioactive compounds (i.e., tannins, saponins, terpenes, flavonoids, and quinones) which provide anthelmintic, antimethanogenic, and antibacterial effects, among other properties [16–18]. Such attributes have motivated the integration of calabash tree into the arboreal stratum of silvopastoral systems, as an interesting component to enhance the establishment of these alternative farming systems in the tropics.

A key challenge in the management of this systems is related to the discrepancy of phenological cycles of the species involved [19]. In addition, the peak of fruit production of calabash tree fruits (CTFs) (wet season) does not match the period of greatest food demand by the herd (dry season). Because of this and the perishable nature of fruits (which deteriorates rapidly under a warm tropical environment due to its high proportion of moisture and soluble carbohydrates), it is necessary to search for feasible alternatives that allows to preserve its nutritional quality from harvest during the wet season until supplementation of cattle in the dry season (around 90 days).

Farmers have empirically resorted to alternatives trying to preserve the fruit from harvesting to supplementation period, based on the local belief that the rigid shell of CTF feature physical conditions to preserve the pulp over time, serving as a "natural mini-silo" [20]. Alternatively, some farmers prefer to conserve the ground fruit into plastic containers adding salt (NaCl) as preservative (i.e., the so-called 'saline silage'; [21]), because due to the low dry matter (DM) content of CTF it could results in spoiled silage, with the risk of developing clostridial organism which thrive in wet conditions [22]. This practice induces an increase in the production costs per kilogram of feed due to the necessity of plastic containers, equipment, NaCl, energy consumption, and additional labor. However, there is a lack of knowledge to confirm either the efficacy of calabash fruit shell ('natural' method) or the effectiveness of silage technique and NaCl addition on calabash fruit conservation.

Therefore, it was hypothesized that the rigid shell of calabash fruit might not be sufficient to preserve the quality of the pulp for a period of 90 days. Furthermore, it was considered that mixing CTF either with its own foliage or other tropical grass hay available on the farm, could increase the DM content of the ensiled mass improving the mixture features and, consequently, the efficiency of conservation (enhancing fermentation profile while reducing gas, fresh and DM losses, and increasing DM recovery), preserving its nutritional quality as a supplement for tropical cattle during the dry season.

Hence, the objective of this research was to compare the effects of using different ensiling treatments on the conservation efficiency, fermentation parameters, and nutritional value of CTF. A secondary objective was to validate the stablished local beliefs that (i) NaCl addition is beneficial for the ensiling process and (ii) the fruit mass may be conserved *per se* in its natural form.

2. Material and Methods

2.1. Experimental Area. The trial was carried out in the commercial farm "El Porvenir," located in the municipality of

TABLE 1: Chemical composition (g/kg of DM) of each ingredient used to perform the experimental treatments.

Item	Ingredient		
	Calabash tree fruit	Calabash tree foliage	Angleton hay
Dry matter[a]	223.3	356.7	941.2
Organic matter	942.7	919.6	916.4
Ash	57.3	80.4	83.6
Crude protein	93.4	121.0	29.8
Total carbohydrates[b]	725.6	784.0	885.4
Neutral detergent fiber	218.8	524.4	758.8
Acid detergent fiber	150.6	356.6	438.1
Hemicellulose	68.2	167.8	320.7
Cellulose	72.3	234.7	365.8
Lignin	78.3	121.9	72.3
Nonfibrous carbohydrates[b]	506.8	259.6	126.6
B1 fraction (starch + pectin)[b]	382.3	228.9	125.6
Water-soluble carbohydrates[c]	124.5	30.7	< 1.0
Ether extract	123.6	14.6	1.2
Gross energy (MJ/kg DM)	18.1	16.4	15.4

Abbreviations: DM, dry matter; FM, fresh matter.
[a]Expressed as g/kg of FM.
[b]Calculated with equations proposed by Sniffen et al. [14].
[c]Determined according to AOAC method 932.12 (°Brix refractometer).

Agustín Codazzi, which belongs to the department of Cesar, in the Colombian Caribbean region (10°60′00″N 73°23′55″ W, altitude 80 m.a.s.l.). The area is located in the tropical dry forest agroecological zone [23]. Weather conditions and soil features were measured during two consecutive years. The average of total annual rainfall was 1300 mm, average relative humidity was 65%, and average annual temperature (T°C) was 28.8°C (min: 24.0°C; max: 34.3°C). The predominant soil type is loamy loam, with pH and organic matter values ranging between 6.5 and 7.0 and 1% and 2%, respectively [12]. The farm accounts for a total area of 182 ha dedicated to extensive livestock production. Of these, 6 ha have been structured in a silvopastoral system with a wide presence of established calabash trees (*Crescentia cujete*) with a 5 m × 5 m-triangular planting frame (~2200 trees in the total area) and an herbaceous stratum based on two native and improved grasses (*Botriochloa pertusa* and *Megathyrsus maximus* cv. Tanzania). The system provides livestock with shade, forages, and fruits, which are used for supplying feed deficit mainly during the dry season. Calabash trees were planted between 2010 and 2012, and since then, they have been regularly producing fruits. A portion of the cattle (around 45 beef cows) has been grazing the area continuously, under a rotational grazing system, from 2014 to date (2025). The farm allocates 25 ha planted with Angleton (*Dichantium aristatum*) to produce hay at the beginning of each dry season. The harvest is at around 45 days after the last grazing, then baled (John Deere R350R; John Deere 449, Moline, IL, United States) and stored indoors in rolls (300 kg each). The remaining area of the farm is intended to be used as pastureland (a silvopastoral system with *Leucaena leucocephala*), timber, and hay production.

2.2. Silage Performing. In order to produce minisilages of CTF, an amount of 1250 fruits (860 kg) was harvested in October 2023. The fruits were transferred to a silage production area, located on the farm, and fully processed using a Trapp hammer grinder (Model TR500, Jaraguá do Sul, SC, Brazil) in order to break the rigid shell of them until obtaining a homogeneous mash with the smallest shell size possible. The experimental treatments were then prepared as described as follows: To evaluate the conservation of the fruit *in natura* (to test the hypothesis that the rigid shell of fruit can preserve the pulp serving as a "natural mini-silo"), a control group (**T0**) was established consisting of six sets of six fruits each (intact, unground, and not ensiled) conserved in sealed polypropylene bags which were stored at ambient T°C in a dry and fresh shed, protected from sunlight, simulating the way local farmers empirically store fruits. To evaluate the calabash fruit conservation as silage, four experimental treatments were evaluated using polyethylene buckets with a capacity of 4 L as experimental minisilos. The treatments were as follows: **T1**, ground whole fruit without any additional additives; **T2**, idem to T1, with the addition of common NaCl at 1.5% of the ensiled mass, simulating the way local farmers empirically ensile fruits; **T3**, a mixture of ground fruits and fresh foliage of the calabash tree in a 30:70 fruit-to-roughage ratio in order to increase DM content of the ensiled mass up to approximately 300 g/kg of FM; and **T4**, a mixture of ground fruits and Angleton hay resulting in a 50:50 fruit-to-roughage ratio aiming to achieve a DM content of the ensiled mass up to approximately 600 g/kg of FM (Figure 1 and Table 2). The two forages were subject to two rounds of shredding through the hammer grinder to ensure a particle size for ensiling of around 1–2 cm. Then, each treatment involving forages was mixed manually by shoveling the material in two 60-kg piles for 10 min each, onto a dry and clean surface to avoid any contamination of the mixture. The two 60-kg piles were then merged and mixed for another 10 min before filling the experimental minisilos.

Samples of each treatment were taken immediately after mixing to represent Day 0. The empty silos (joint bucket and lid) were weighed and immediately the bucket dimensions (height and diameter in the basis and the top) were measured to calculate the volume occupied by the ensiled mass. Then, they were filled according to the experimental treatments, taking care in T1 and T2 to leave a 5-cm gap between the top of the mixture and the silo opening edge, to prevent them from exploding due to the internal gas pressure. In those silos filled with T3 and T4, the material was placed in layers of ± 5 cm which were manually compacted using a blunt wooden log until the mixture showed signs of not compacting further. The same procedure was repeated layer after layer until the ensiled mass reached 1 cm above the level of the edge at the top of the silo. Finally, each silo was immediately sealed with a snap-on lid and adhesive tape, weighed (as well as the bags in T0) and stored in a safe place. Data regarding the dimensions of buckets and the

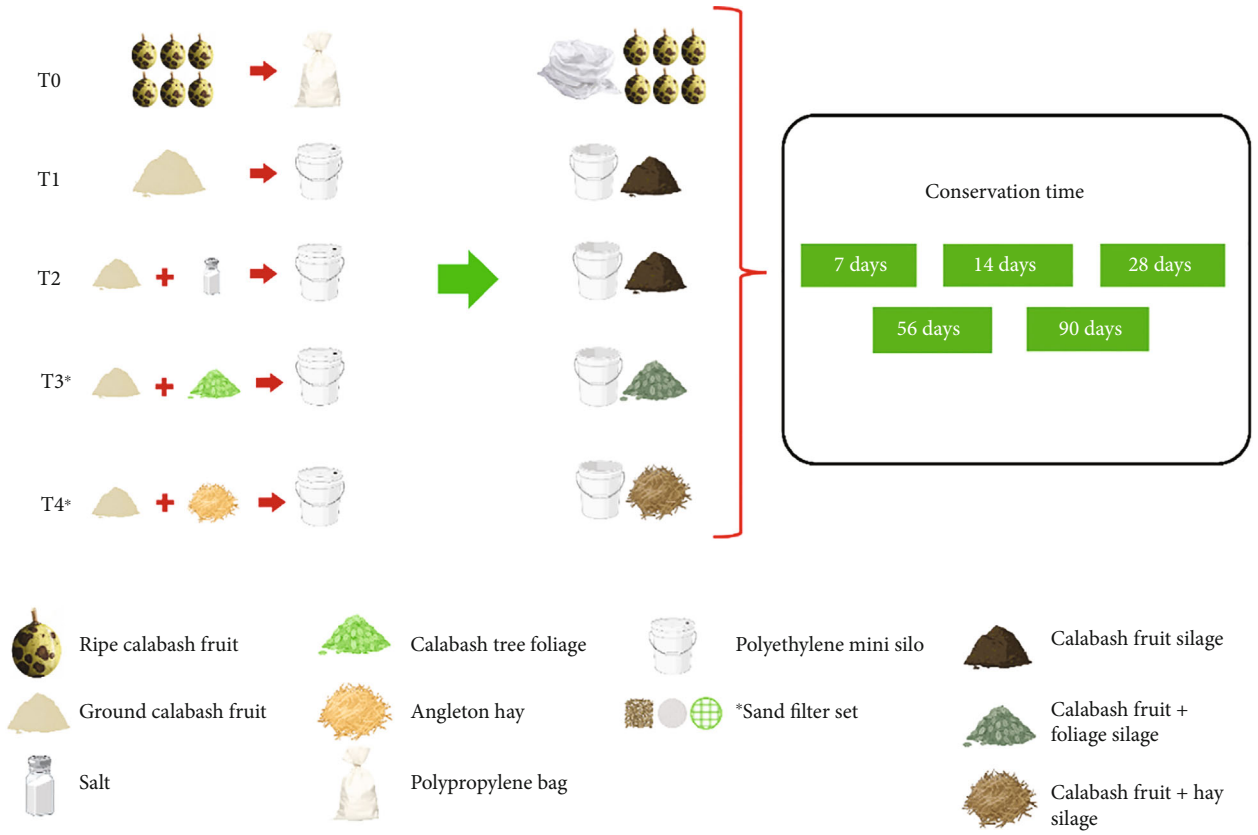

FIGURE 1: Experimental design scheme indicating the components used in each treatment and the conservation times (sampling). It had been used 25 replicates per treatment (five replicates for each conservation time). Created in BioRender. Rojas-Meza, D.A. (2025).

TABLE 2: Initial ensiled mass, dry matter content, and calculated specific density of experimental treatments.

Variable	Ensiled treatments			
	T1	T2	T3	T4
Initial dry matter (g/kg FM)	204.0	209.5	316.7	582.3
Average ensiled mass (kg/mini-silo)	2.75 ± 0.1	2.76 ± 0.1	2.28 ± 0.2	1.04 ± 0.1
Specific density (kg FM/m^3)	925 ± 45	946 ± 53	630 ± 68	264 ± 37
Specific density (kg DM/m^3)	211 ± 13	228 ± 13	191 ± 22	156 ± 21

Abbreviations: **T1**, ground fruit without any additional additives; **T2**, ground fruit +1.5% of salt (saline silage); **T3**, mixture of ground fruits and foliage of calabash tree in a 30:70 fruit-to-roughage ratio; **T4**, mixture of ground calabash tree fruits and Angleton hay in a 50:50 fruit-to-roughage ratio.

weighed ensiled mass (this latter calculated as the total weight of the sealed silo minus the weight of the empty silo) were used to calculate the initial specific density.

As an experimental design, bags and silos were distributed in a completely randomized design following a 5 × 5 factorial scheme with five experimental treatments (T0, T1, T2, T3, and T4) and five conservation times (7, 14, 28, 56, and 90 days) with five replicates for a total of 125 experimental units (Figure 1).

On October 21, all the experimental units (25 bags and 100 minisilos) were transferred to the facilities of the *Laboratorio de Biotecnología Ruminal* at the *Universidad Nacional de Colombia* (Medellín headquarter) and stored in a cool and dry place at ambient T°C during the conservation periods, where they were subsequently opened (according to the conservation times) and analyzed for the parameters of fermentation, conservation, and nutritional quality.

2.3. Sampling and Laboratorial Analyses. For each conservation time, both bags and minisilos were opened in accordance with a standardised procedure: Prior to opening, each replicate was weighed on a digital balance; fruits were opened using a handsaw whereas the silos were opened by removing the adhesive tape and lid. The fresh matter losses (FMLs), gas losses (GLs), and DM recovery (DMR) were calculated following the procedures described by Nascimento et al. [24].

T°C inside the fruits and silage was measured using a digital penetration thermometer (Model Checktemp HI98501, Hanna Instruments, Padova, Italy). When either

fruits' pulp or silage showed some macroscopic signs of apparent fungal or yeast presence (determined visually by the presence/absence of either mold and/or colonies with a creamy consistence), the affected material was separated, weighed to calculate the FML, and then discarded. The pulp was removed from the shell, which was weighed afterwards to be excluded from the initial weight, and pulp from all fruits was manually mixed into a polyethylene bowl until a soft and homogeneous mash was achieved, in order to obtain a compound (representative) sample from each experimental unit. In the silos, the top layer of silage (1–2 cm in depth) was removed and two representative samples (~300 g) were collected from all the experimental treatments. The first one was stored at −18°C for further chemical analyses. The other one was used to extract a few drops of liquid through manual pressure, which was used to determine the brix degrees using a portable refractometer (Brixco, Model 3095 equipped with a 0%–20% scale) in order to calculate the concentration of soluble carbohydrates (SC; AOAC, 932.12) according to Horwitz [25]. The rest of the material (of the second sample) was dried in a forced air circulation oven at 55°C for 72 h, ground in a Willey mill (Thomas Scientific, Swedesboro, NJ, United States) using a 1-mm sieve and analyzed for DM (AOAC, 934.01), ash (AOAC, 942.05), and EE (AOAC, 920.39) according to Lee [26]. The TN was determined by distillation using a Kjeldahl equipment according to Latimer [27] and CP was calculated as TN × 6.25. Gross energy (GE) was obtained by combusting samples in a calorimeter bomb (Parr Instrument Company Model 1341, Moline, IL, United States). The neutral detergent fiber(NDF) and acid detergent fiber (ADF) were sequentially determined in an ANKOM^{200} Fiber Analyzer (ANKOM Technology Corporation, Fairport, NY, United States). The NDF content and its components (cellulose, hemicellulose, and lignin) were analyzed according to Van Soest et al. [28], while total carbohydrates (TCs), nonfibrous carbohydrates (NFCs), and B1 fraction were calculated according to Sniffen et al. [14] (Table 1).

From the stored samples of each silage (−18°C), 25 g was used for performing a liquid extraction following the procedures described by Rossi et al. [29]. The liquid extract was analyzed for ammonia nitrogen (NH_3-N) by distillation according to Fenner [30] and organic acids (lactic, acetic, and propionic) using a high-performance liquid chromatograph (HPLC; Agilent model 1100 Series, Agilent, CA, United States) equipped with a UV/VIS detection system and an apolar column (Supelcogel H, 7.8 mm × 300 mm). The polar mobile phase consisted of a 0.1% buffer phosphate solution. The presence of the acids was detected by UV absorbance, at a wavelength of 210 nm, while a second 25-g sample was used for liquid extraction and analysis of buffering capacity (BC) using the titration technique according to Playne and McDonald [31]. The GL, NH_3-N, and the remaining parameters were not assessed on treatment T0 due to its specific storage conditions (not silage process).

2.4. Statistical Analysis. Parameters related to fermentation profile (pH, T°C, SC and NH_3-N concentration, BC, and organic acid concentration) and conservation efficiency (silage density, FML, GL, and DMR) data were analyzed by ANOVA as a completely randomized design, following a 5 × 5 factorial scheme with five treatments and five conservation times. Treatment and time effects were considered as fixed effects in the model.

When the ANOVA result was significant, a post hoc LSD test with Holm multiplicity adjustment was used to compare treatment means, conservation time means or the first-order treatment × time interaction. The significance level of 5% was used for all tests.

A desirability function approach (Harrington, [32, 33]) was used to assess the efficiency of conservation parameters and fermentative characteristics, in order to define which was the best treatment. The desirability index (*D*) consists of a weighted and objective rating given to a product or process within a strictly ordinal scale established between 0 and 1, in which a lower index corresponds to a less desirable result and a higher one to a more desirable result, the latter being the one that most resembles a previously established ideal (or desirable as the name of the index suggests). The use of these continuous and discontinuous desirability functions proposed by Derringer and Suich [34] is very useful in cases such as the present study, in which the objective is to determine precisely (avoiding subjectivities) and based on a reference (ideal) which is the best product or process among multiple options (experimental treatments) that are the result of the interaction of various fermentation and quality parameters with different degrees of influence on the final result. For this purpose, a *D* calculation matrix was proposed, based on 10 parameters (DMR, lactic acid concentration, pH, MLF, BC, NH_3-N concentration, SC concentration, GL, acetic acid concentration, and lactic-to-acetic ratio) and their influence on the final product was assessed on a scale of 1 to 5 where 1 = *very low*, 2 = *low*, 3 = *medium*, 4 = *high*, and 5 = *very high*. Thus, the lactic acid concentration, pH, and DMR variables were assigned a score of 5, followed by FML, BC, and NH_3-N with a score of 4 and the remaining parameters with a score of 3, following the criteria suggested by Cherney and Cherney [35] for estimating the quality of fermentation and its influence on the efficiency of conservation and quality of silages. The "desired" limits for each parameter were defined according to Kung Jr., et al. [36] considering as "ideal" a 30%–40% of DM corn silage, since it has been the feed that typically shows the most desirable ensilability features. The functions proposed by Derringer and Suich also allow penalizing the partial index (*d*) of each weighting parameter in three different scenarios: when a maximum value of this parameter is expected, when a minimum value is expected, and when reference values are expected in a specific range. Based on the above, minimum values were desirable for parameters such as GL, MLF, pH, NH3-N, and BC, and the higher and further from the ideal, the lower the *d* for that parameter. The opposite was true for parameters such as DMR, where the highest possible values are expected, and the higher the value, the better the *d*. For the remaining variables, ranges were established as desirable, and values above or below this range were penalized with a lower *d*. Finally, the *D* for each treatment evaluated at each storage time was calculated as the weighted average of the

partial desirability values exhibited for each parameter. The data were analyzed using the 'indes' customised function developed by Correa-Londoño [37].

All statistical analyses were performed using R Studio Software version 3.6.0 (2024; R Core Team) after testing the mathematical assumptions of the model (Shapiro–Wilk and Levene tests) and those from the desirability function approach. When the dataset did not meet the mathematical assumptions of the model, Box–Cox family transformations were applied.

3. Results and Discussion

3.1. DM Content and Specific Density of Silages Including Tropical Forages. The initial DM concentration, ensiled mass per silo (average), and the calculated specific density per treatment are shown in Table 2. Treatments with forage in the ensiled mass increased their initial DM content while the average amount of ensiled mass and their specific density were reduced, when compared to those without forage inclusion.

The inclusion of tropical forages to silage in treatments T3 and T4 increased the DM content by 55% and 185% and NDF content by 111% and 201%, respectively, compared to treatment T1. This increase is attributed to the higher content of DM and NDF of calabash tree foliage with respect to CTF (Table 1), which led to a more voluminous mixture with lower specific density and less average weight of ensiled mass/minisilo with T4 reporting the lowest values in those parameters, followed by T3 (Table 2).

DM and NDF content are considered important parameters in silage production since these, along with particle size, have a high negative correlation with compaction capacity, which consequently determines the specific density, fermentative profile, nutritional quality and conservation success of silages [38–40]. From a practical perspective, the specific density is important as it defines the physical space and inputs required to conserve a certain amount of silage (kg FM or DM/m^3), which also defines part of the total storage costs [41, 42].

Commonly, silages based on forages with a DM content between 30% and 40% exhibit appropriate specific density and ideal fermentation parameters that guarantee the conservation of the ensiled mass over time. The DM content below or above this range usually affects fermentation parameters, which could increase the risk of losses, affecting conservation efficiency and nutritional quality of silage ([36] as will be discussed later.

3.2. Conservation Process and Changes on Chemical Composition. Table 3 shows a comparison between initial and final chemical composition per treatment after 90 days. A decrease in CP, EE, and NFC which reflected on DM, OM, and GE composition in treatments T1, T2, and T3 was reported, whereas T0 and T4 showed a dramatic decrease in all the nutritive fractions. On the contrary, hemicellulose, cellulose, and lignin, and consequently, NDF and ADF increased in T1, T2, and T3 at the end of the conservation period, as well as the ash fraction in these treatments.

Results on the chemical composition of treatment T0 (no silage) reported excessive losses in all nutritional fractions that dramatically compromised its nutritional value (Table 3). The macroscopic presence of fungus (mold), as well as damage by insects and other unidentified organisms that promoted the CTF pulp decomposition, was observed from 28 to 90 conservation days (Supporting Information (available here)). This could be attributed to the fact that the shell, despite its physical characteristics, constitutes a living and selectively permeable tissue, which allows the exchange of gases and other substances with the surrounding environment [43].

At the end of the conservation period, an apparent increase of CP and EE concentrations (expressed as % of final DM, from 9.3% to 22.7% and 12.4% to 22.8%, respectively—data not shown) was observed. However, this nutrient increase was fictional, but rather a concentration effect of these nutritive fractions caused by the protection of pericarp and seed coat to the endocarp of seeds (structure particularly rich in CP and EE), avoiding them from suffering high losses as occurred with the pulp constituents. Due to the excessive loss of DM in treatment T0, the remaining CP and EE fractions become a major portion in the composition of a decreased final DM, causing the concentration effect described above.

As expected, all the ensiled treatments (T1 to T4) reported a decrease in NFC, CP, and EE as a consequence of the fermentative process after 90 conservation days, which also led to a concomitant decrease in DM, OM, and GE. Silage is a long-recognized conservation technique, used to conserve the nutritional quality of feed resources mass over time. However, it involves changes in the nutritive composition and some losses related to the microbiological and biochemical activity of the conservation process [36]. In silages, nutritive fractions such as NFC, CP, and EE are expected to be used for microbial growth during the phases prior to their stabilization [44], which results in the synthesis of organic acids (lactic and acetic acids, necessary for an efficient ensiled mass conservation), some alcohols, esters, and nitrogen compounds such as NH_3-N coming from the decomposition of complex nitrogenated molecules [45]. The consumption of these nutrients by silage microorganisms is also externally reflected in the production of gases, effluents and a concomitant shrinking of silage mass [22].

In contrast, an increase in the cell wall components (cellulose, hemicellulose, and lignin) in the final chemical composition of silages was observed in our study. This is related to a concentration effect [46–48] caused by the decrease in NFC from the activity of the microorganism populations, that dispense with the consumption of the fibrous fraction since these nutrients are not considered a main source of energy for cell growth in the presence of reasonable amounts of soluble nutrients such as those mentioned initially [22, 45]. The latter is then reflected in the proportional increase of the fibrous fraction in the final DM of silages (Table 3). The addition of NaCl at 1.5% of FM in treatment T2 was also reflected in the increase of the mineral fraction (Table 3) compared with T1, which was expected since common NaCl (composed of chlorine

TABLE 3: Initial (0 days) and final (90 days) chemical composition for each experimental treatment. Data of initial and final time are expressed in grams (absolute quantity) and differences (Dif) are expressed in percentages.

Nutrients (g)	No silage			Ensiled treatments											
	T0			T1			T2			T3			T4		
	Initial	Final	Dif	Initial	Final	Dif	Initial	Final	Dif	Initial	Final	Dif	Initial	Final	Dif
Dry matter[a]	702.5	215.1	−69.4	638.7	537.0	−15.9	700.7	570.3	−18.6	708.1	623.0	−12.0	587.1	302.1	−48.5
Organic matter	662.2	197.9	−70.1	599.9	493.2	−17.8	643.9	467.1	−27.4	654.7	566.3	−13.5	541.0	279.6	−48.3
Crude protein	65.6	48.8	−25.6	57.0	58.6	2.9	65.5	60.6	−7.5	81.5	77.4	−5.1	24.6	11.0	−55.4
Total carbohydrates	509.7	100.0	−80.4	465.8	375.4	−19.4	491.8	346.3	−29.6	546.5	471.6	−13.7	501.8	260.8	−48.0
Neutral detergent fiber	153.7	97.0	−36.9	139.0	186.4	34.1	153.3	188.9	23.2	325.6	362.7	11.4	384.7	227.2	−40.9
Acid detergent fiber	105.8	73.6	−30.4	94.8	146.3	54.3	105.5	136.4	29.2	221.7	253.6	14.4	224.8	140.1	−37.7
Hemicellulose	47.9	23.4	−51.1	44.2	40.2	−9.0	47.8	52.6	10.0	103.9	109.1	5.0	159.9	87.2	−45.5
Cellulose	50.8	49.9	−1.7	58.2	76.9	32.2	50.7	73.5	45.1	141.9	164.7	16.1	181.7	111.0	−38.9
Lignin	55.0	23.7	−56.9	36.6	69.3	89.4	54.9	62.8	14.5	79.8	88.9	11.4	43.1	29.0	−32.7
Nonfibrous carbohydrates	356.0	3.0	−99.2	326.9	188.9	−42.2	338.4	157.4	−53.5	220.9	108.9	−50.7	117.1	33.6	−71.3
B1 fraction	267.5	2.8	−99.0	247.6	146.2	−41.0	248.7	105.8	−57.5	165.2	74.8	−54.7	86.8	24.7	−71.5
Soluble carbohydrates	88.5	0.2	−99.8	79.2	42.7	−46.1	89.8	51.6	−42.5	55.7	34.1	−38.7	30.4	8.9	−70.7
Ether extract	86.9	49.0	−43.6	77.1	59.2	−23.2	86.6	60.3	−30.4	26.7	17.3	−35.2	14.5	7.8	−46.1
Ash	40.2	17.3	−57.1	38.9	43.8	12.8	56.8	103.2	81.5	53.5	56.7	6.1	46.1	22.5	−51.2
Gross energy (MJ)	12.7	5.1	−59.9	11.5	10.7	−7.0	12.7	10.1	−20.5	11.9	11.4	−4.2	9.3	5.2	−44.1

Note: Values in the "Dif" column accompanied by a negative sign express a percentual decrease of the nutrient and values without a negative sign (positive value) indicate a percentual increase of the nutrient in the total composition.

Abbreviations: **T0**, ripe calabash tree fruit (unground and not ensiled); **T1**, ground fruit without any additional additives; **T2**, ground fruit +1.5% of salt (saline silage); **T3**, mixture of ground fruits and foliage of calabash tree in a 30:70 fruit-to-roughage ratio; **T4**, mixture of ground calabash tree fruits and Angleton hay in a 50:50 fruit-to-roughage ratio.

[a]Expressed as g/kg of FM.

and sodium) is considered an important source of additional minerals.

Despite a descriptive approach of data in Table 3 and contrary to what was expected, NaCl addition appeared to have no positive effect on the conservation of nutritive value of CTF after a 90-day period. NaCl addition is one of the most traditional methods for inhibiting the growth and survival of undesirable spoilage bacteria and fungi naturally present in foods, favoring the growth of more NaCl-tolerant organisms such as lactic acid bacteria (LAB) [49]. However, treatment T2 showed a higher decrease in NFC, CP, and EE and, consequently, a lower amount of final DM, OM, and GE compared with treatment T1 (control without NaCl addition). This could be attributed to the hygroscopic nature of NaCl which helps to draw water and sugars out of plant tissues during the fermentative conservation process, filling any air pores present in the ensiled mass. Then, it may finally result in reduced oxygen conditions that promote a higher rate of the fermentation process [21], and the major consumption of the nutritional fractions potentially used by microorganisms, reflected also in higher GLs (Table 4 and Figure 2) as reported in this study.

Regarding the changes in chemical composition in treatment T3 and T4, the first one reported a similar behavior to treatments T1 and T2, showing a decrease in NFC, CP, and EE with a concentration effect on ash content and cell wall components after a 90-day conservation period. Treatment T4 showed a decrease in all the nutritive fractions and performed with a similar trend to that previously reported in treatment T0, although it could be noticed that losses were lower in T4 than T0 (Table 3).

Treatment T3 showed a trend to lower decrease in the soluble nutrients previously mentioned, as well as a lower decrease in DM, OM, and GE, compared to silages that did not include forage in the mixture, with the exception of similar NFC (Table 3). This could be influenced by its lower moisture and SC content, as a positive result of the roughage inclusion. As described by Nascimento et al. [24], according to several studies the ideal moisture and SC concentrations in silages range between 60% and 70% and 6% and 8%, respectively, which determine adequate fermentation processes and conservation features as shown in this treatment. Such ideal conditions do not allow the development of a certain group of microorganisms, which under excessive moisture and SC contents are typically responsible for an increased consumption of soluble nutrients such as CP and EE [50, 51] as has been reported in treatments T1 and T2.

3.3. Fermentation Parameters and Conservation Efficiency. Differences between the fermentative parameters after 90 conservation days and kinetics of fermentation parameters along the five assessed conservation times are shown in Table 4 and Figures 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, and 12, respectively. There was a significant effect of treatment, and conservation time on all parameters, as well as for the interaction treatment × conservation time with the exception of NH_3-N, acetic acid concentration, and lactic-to-acetic ratio.

TABLE 4: Fermentation parameters of experimental treatments after 90 days conservation period.

Item	No silage	Ensiled treatments					*p* value		
	T0	T1	T2	T3	T4	SEM	*T*	*t*	*T* ∗ *t*
FM losses (% of FM)	81.26^{a}	2.59^{b}	1.92^{b}	2.18^{b}	42.97^{c}	6.95	< 0.001	0.004	0.037
Gas losses (% DM)	NA	2.47^{a}	2.99^{b}	2.12^{a}	3.11^{b}	0.30	< 0.001	< 0.001	< 0.001
DM recovery (%)	30.75^{a}	84.16^{b}	81.42^{b}	88.16^{b}	50.10^{c}	2.39	< 0.001	< 0.001	< 0.001
Temperature (°C)	23.17^{a}	23.66^{b}	23.68^{b}	24.12^{c}	25.06^{d}	0.15	< 0.001	< 0.001	< 0.001
Initial SC (%)	12.40^{a}	11.40^{b}	13.40^{a}	7.75^{c}	5.20^{d}	0.49	< 0.001	< 0.001	0.018
Final SC (%)	<0.1^{a}	7.95^{b}	9.05^{b}	5.48^{c}	2.94^{d}	0.54	< 0.001	< 0.001	0.018
pH	6.68^{a}	3.75^{b}	3.78^{b}	4.45^{c}	5.01^{d}	0.06	< 0.001	< 0.001	< 0.001
NH_3-N (% of total N)	NA	12.79^{a}	12.26^{a}	7.82^{b}	9.56^{b}	1.05	< 0.001	0.007	0.784
Buffer capacity (mE/100 g of DM)	NA	75.59^{a}	54.74^{b}	44.20^{c}	14.21^{d}	3.17	< 0.001	< 0.001	< 0.001
Lactic acid (% DM)	NA	8.70^{a}	6.70^{a}	3.28^{b}	1.57^{c}	0.74	< 0.001	< 0.001	< 0.001
Acetic acid (% DM)	NA	2.84^{a}	1.41^{b}	1.66^{b}	0.71^{c}	0.22	< 0.001	<0.001	0.223
Propionic acid (% DM)	NA	ND	ND	ND	ND	—	—	—	—
Lactic:acetic	NA	3.07^{a}	4.88^{b}	1.95^{c}	2.22^{ac}	0.29	<0.001	<0.001	0.087

Note: Estimated marginal means (**EMM**) per treatment followed by different superscripted letters in the same row differ ($p < 0.05$).
Abbreviations: **DM**, dry matter; **FM**; fresh matter; **NA**, not applicable; **ND**: not detected; **NH_3-N**, ammonia nitrogen; **SC**, soluble carbohydrates; **SEM**, standard error of the mean; ***T***, experimental treatment; ***t***, conservation time; ***T* ∗ *t***, first-order interaction of experimental treatment × conservation time; **T0**, ripe calabash tree fruit (unground and not ensiled); **T1**, ground fruit without any additional additives; **T2**, ground fruit +1.5% of salt (saline silage); **T3**, mixture of ground fruits and foliage of calabash tree in a 30:70 fruit-to-roughage ratio; **T4**, mixture of ground calabash tree fruits and angleton hay in a 50:50 fruit-to-roughage ratio.

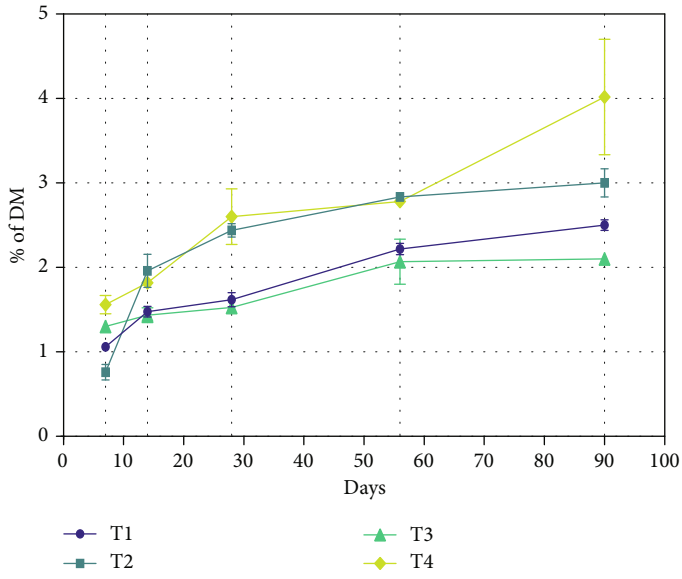

FIGURE 2: Gas losses; kinetics by experimental treatment ($p < 0.001$) over a conservation period of 90 days. Conservation time effect ($p < 0.001$) assessed at 7, 14, 28, 56, and 90 days. Interaction of treatment × conservation time ($p < 0.001$). T0 not assessed.

No silage treatment (T0) showed the worst conservation parameters with the highest FML and SC consumption, poorest DMR, and a close to neutral pH final value, when compared with silage treatments (T1 to T4). According to all these findings, it can be concluded that the hard shell of CTF is not sufficient to protect the pulp and to promote anaerobic conditions for an acid lactic fermentation as expected in silage technique, and contrary to the local belief of technicians and smallholders, it does not behave as a "natural mini-silo." This is a very important aspect to consider as, in silages, pH values higher than 5 could allow proliferation of undesirable clostridial, enterobacteria and other microorganisms such as fungus that deteriorate the quality of ensiled mass [22, 36], thus compromising animal health and performance.

Such bad results invalidate the T0 as a potential method for CTF conservation for more than 28 days, emphasizing that DMR reported significant losses (21% at 28 conservation days; Figure 4) which exceed the maximum expected of 10%–15%. Those findings strengthen the necessity of

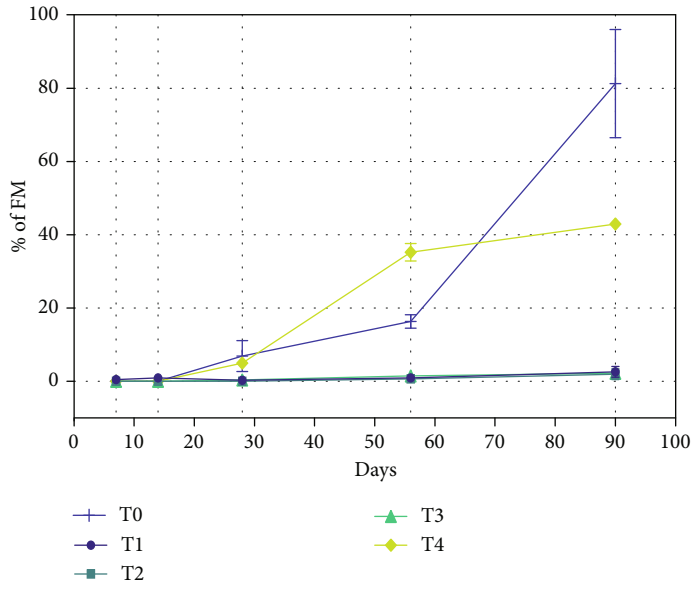

Figure 3: Fresh matter losses; kinetics by experimental treatment ($p < 0.001$) over a conservation period of 90 days. Conservation time effect ($p = 0.004$) assessed at 7, 14, 28, 56, and 90 days. Interaction of treatment × conservation time ($p = 0.037$).

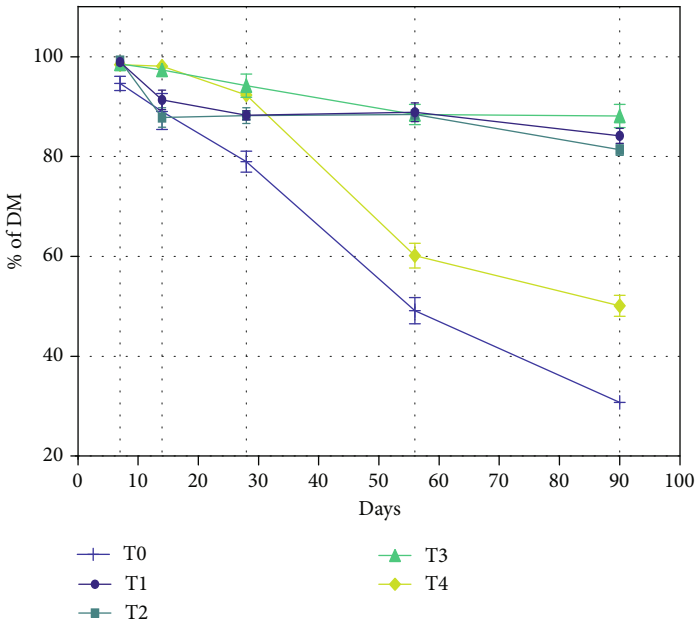

Figure 4: Dry matter recovery; kinetics by experimental treatment ($p < 0.001$) over a conservation period of 90 days. Conservation time effect ($p < 0.001$) assessed at 7, 14, 28, 56, and 90 days. Interaction of treatment × conservation time ($p < 0.001$).

employing containers of synthetic material since its resistance to environmental exposure promotes anaerobic conditions for adequate fermentative conservation over time.

Ensiled treatments T1, T2, and T3 showed significant differences ($p < 0.001$) in FML and DMR compared to T0 and T4, without significant differences between them. Treatments T2 and T4 reported the higher GL values which differed statistically ($p < 0.001$) from T1 and T3 which showed the lowest ones. The positive effect of improved levels on moisture and SC could also be evidenced in the lower FML and GL; since the fewer fermentative losses without compromising fermentation parameters and conservation, the more efficient this process will be [24, 35].

It should be noted that T4 exhibited elevated pH and FML (4.74% and 42.6%, respectively) after 90 days of conservation. As previously described in Section 3.1, the higher decrease in all the nutritive fractions observed in T4 can be linked to the excessive inclusion of Angleton hay in the fruit (i.e., roughage silage ratio), resulting in an excessive increase of DM and fibrous fraction thus dramatically reducing its specific density and average amount of ensiled mass/minisilo (Table 2). Consequently, compactness was also affected

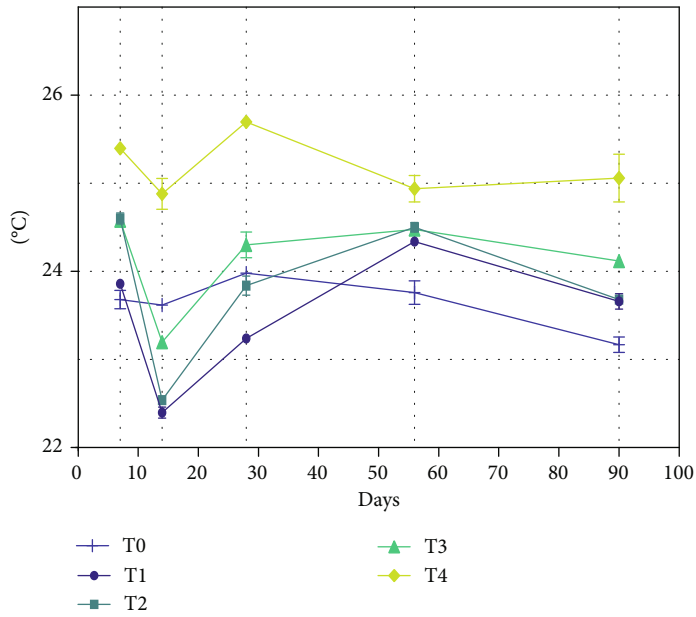

Figure 5: Temperature of ensiled mass; kinetics by experimental treatment ($p < 0.001$) over a conservation period of 90 days. Conservation time effect ($p < 0.001$) assessed at 7, 14, 28, 56, and 90 days. Interaction of treatment × conservation time ($p < 0.001$).

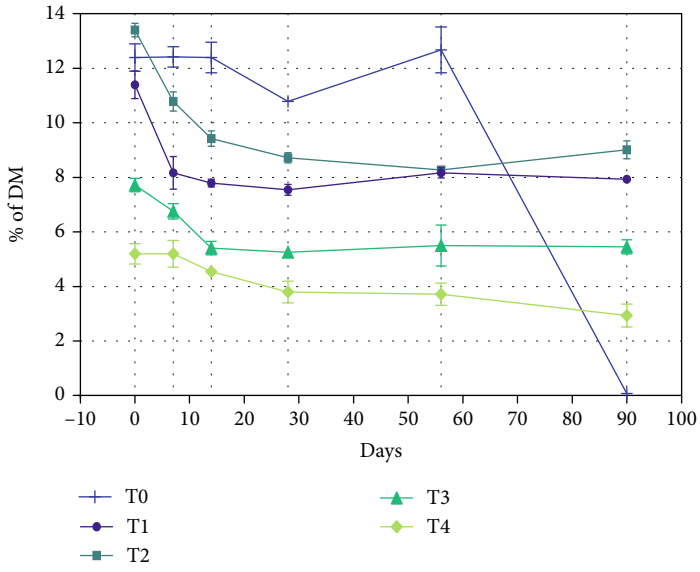

Figure 6: Soluble carbohydrates concentration; kinetics by experimental treatment ($p < 0.001$) over a conservation period of 90 days. Conservation time effect ($p < 0.001$) assessed at 7, 14, 28, 56, and 90 days. Interaction of treatment × conservation time ($p = 0.018$).

causing a "spring effect" in the ensiled mass that impeded a proper extraction of oxygen from silage during the conservation process. This, besides the lowest SC concentration observed in this treatment compared to others (Table 4; Figure 6), impeded the drop of pH as expected (Figure 7). The last is also likely related to the poorest organic acid synthesis in this treatment (Figures 10 and 11), which allowed fungus development inside the minisilos, as evidenced in its opening from 28 until 90 conservation days (Supporting Information). The presence of fungus resulted in FM losses of 35% and 43% at 56 and 90 conservation days, respectively (Table 4 and Figure 3), dramatically affecting the DMR rate (Table 4 and Figure 4) and causing a large loss of nutrients (Table 3). Several studies [36, 52–54] suggest that, when the DM of the ensiled mass exceeds 45%, pH is negatively affected since metabolic water, necessary for LAB growth, is reduced as the DM content increases, thus reducing the necessary organic acids concentrations to prevent the undesirable growth of lactate (assimilating yeasts and fungi responsible for silage spoilage), as reported in this study.

Further studies should be conducted in order to achieve the most appropriate fruit to roughage ratio in treatment T4, allowing for better compaction, increasing the specific density and SC concentration, which will reflect in a better

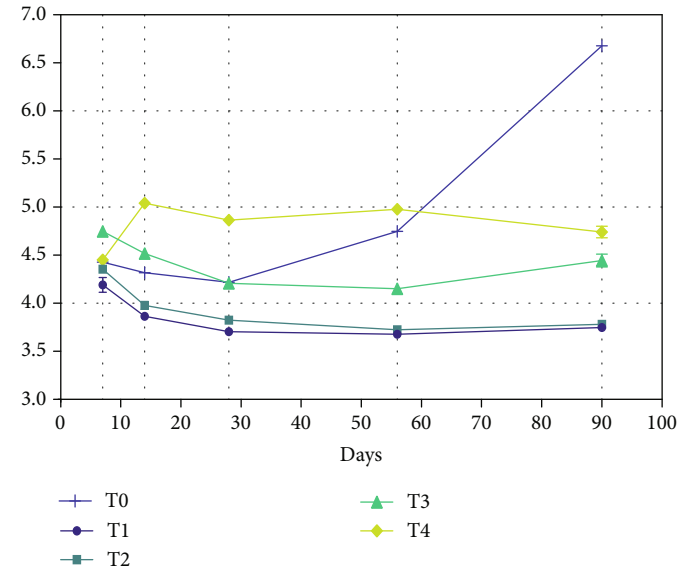

FIGURE 7: pH; kinetics by experimental treatment ($p < 0.001$) over a conservation period of 90 days. Conservation time effect ($p < 0.001$) assessed at 7, 14, 28, 56 and 90 days. Interaction of treatment × conservation time ($p < 0.001$).

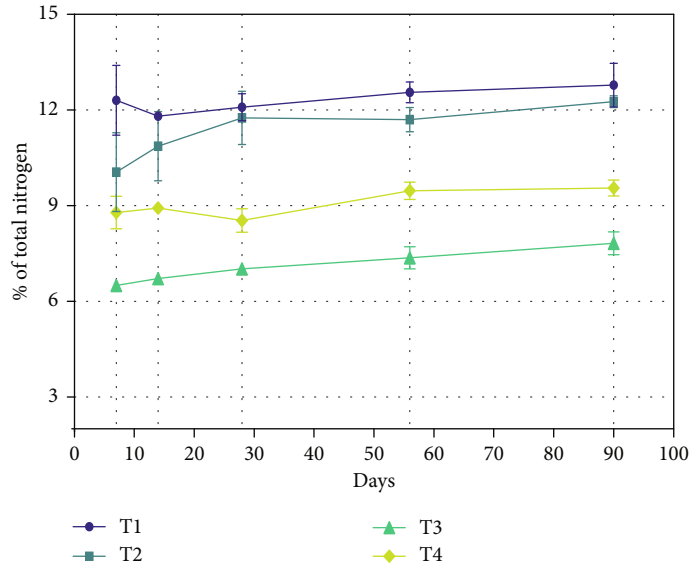

FIGURE 8: Ammoniacal nitrogen concentration; kinetics by experimental treatment ($p < 0.001$) over a conservation period of 90 days. Conservation time effect ($p = 0.007$) assessed at 7, 14, 28, 56, and 90 days. Interaction of treatment × conservation time ($p = 0.784$). T0 not assessed by ammoniacal nitrogen concentration.

fermentation profile, lower losses and better nutritional value of silage for livestock feeding purposes [42, 55].

The addition of NaCl at 1.5% in T2 did not have a significant effect on FML, DMR, T°C of ensiled mass, pH, and final SC and NH_3-N concentrations when compared to T1 after 90 days (Table 4). However, an increase in GL ($p < 0.001$) and a reduction in acetic acid concentration ($p < 0.001$) and BC ($p < 0.001$) were observed.

Although the addition of NaCl could stimulate the growth and activity of LAB as previously mentioned, it should be noted that this effect is dose-dependent. In a study conducted by Yang et al. [56], to evaluate the fermentative activity of Northeast-style sauerkraut with different NaCl concentrations, it was reported that 0.5% NaCl concentration stimulated the growth and activity of *Lactobacillus*, while its population was reduced in the presence of higher NaCl concentrations (1.5%, 2.5%, and 3.5%), suggesting that levels equal to or greater than 1.5% affect LAB populations. Huang et al. [57], evaluating acid production during fermentation of high-NaCl kitchen wastewater, reported a reduction in acid production by LAB with an increase in NaCl concentration from 0 to 8 g/L. These results demonstrated that although optimal Na+ levels improve enzymatic activity and osmotic balance, high concentrations can inhibit LAB

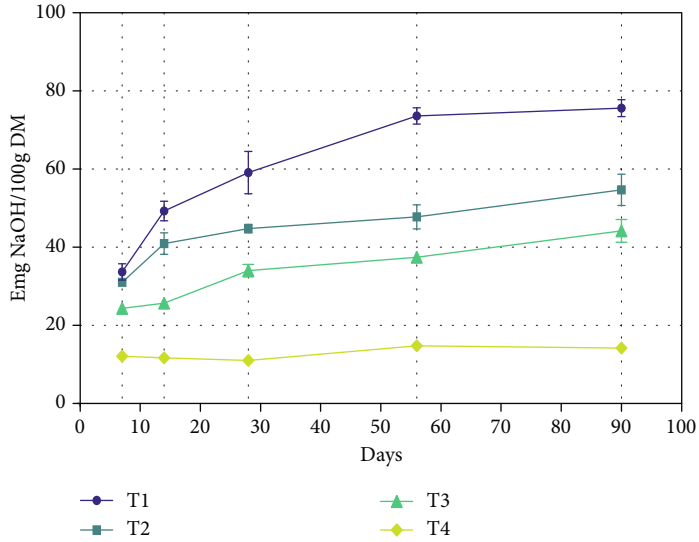

Figure 9: Buffering capacity; kinetics by experimental treatment ($p < 0.001$) over a conservation period of 90 days. Conservation time effect ($p < 0.001$) assessed at 7, 14, 28, 56, and 90 days. Interaction of treatment × conservation time ($p < 0.001$). T0 not assessed by buffering capacity.

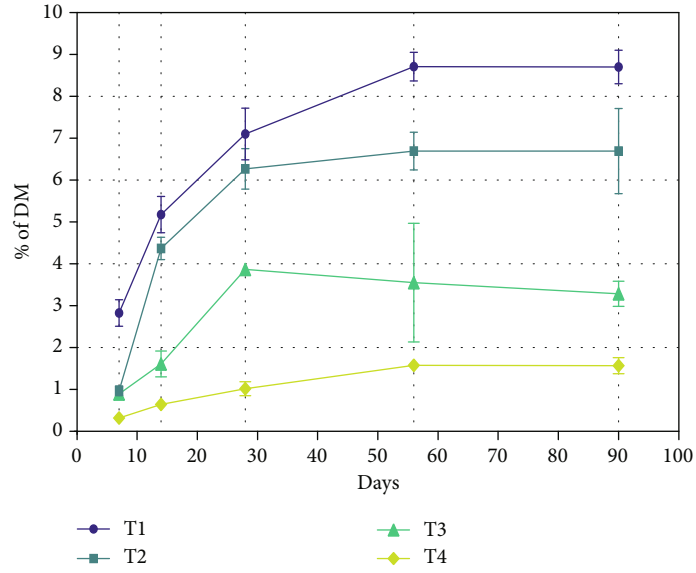

Figure 10: Lactic acid concentrations; kinetics by experimental treatment ($p < 0.001$) over a conservation period of 90 days. Conservation time effect ($p < 0.001$) assessed at 7, 14, 28, 56, and 90 days. Interaction of treatment × conservation time ($p < 0.001$). T0 not assessed.

growth and fermentative function, probably allowing the appearance of salinity-resistant microorganisms. These reports could probably explain either the excessive consumption of the soluble nutritive fractions (Table 3) and higher GL (Table 4 and Figure 2) caused by the activity of undesirable salinity-resistant microorganisms, as well as the lower concentrations of lactic and acetic acid (Table 4 and Figures 10 and 11), due to the inhibition of LAB activity by NaCl addition.

Several authors [22, 36, 45] reported the activity of undesirable microorganisms present in the fermentation process that can consume part of lactic acid, thus converting it to other acids, alcohols, esters, CO_2 and water, these being responsible for the increased consumption of nutrient fractions and losses due to gas production. This could also explain the results reported in this study related to treatment T2. However, further studies including analysis to confirm the presence of alcohols, esters and the assessment of changes in microbial populations, should be conducted in order to allow us to confirm such hypotheses.

It is unclear whether the decrease of BC values in T2 could be motivated by NaCl addition. According to Pahlow et al. [22], BC is dependent mainly on cation exchange capacity (CEC) of the fiber and on the fermentation of protein to NH_3-N. Considering that there was no significant difference in NH_3-N concentrations between treatments T1

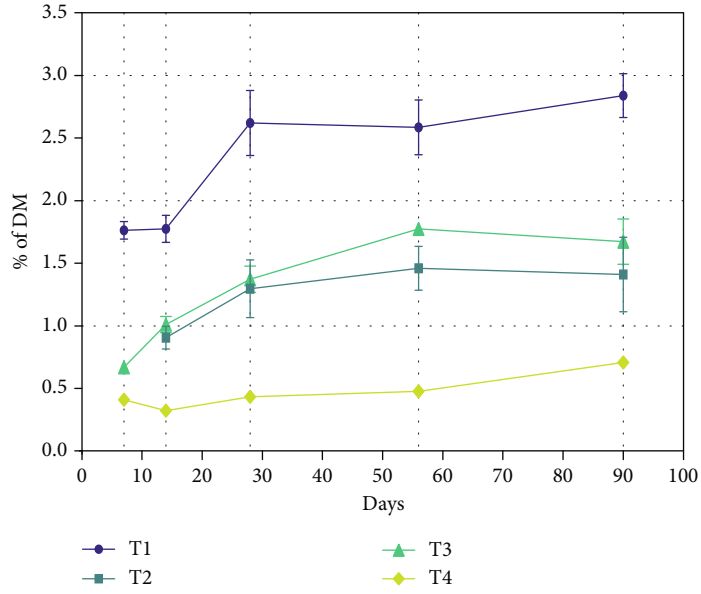

Figure 11: Acetic acid concentrations; kinetics by experimental treatment ($p < 0.001$) over a conservation period of 90 days. Conservation time effect ($p < 0.001$) assessed at 7, 14, 28, 56, and 90 days. Interaction of treatment × conservation time ($p = 0.223$). T0 not assessed.

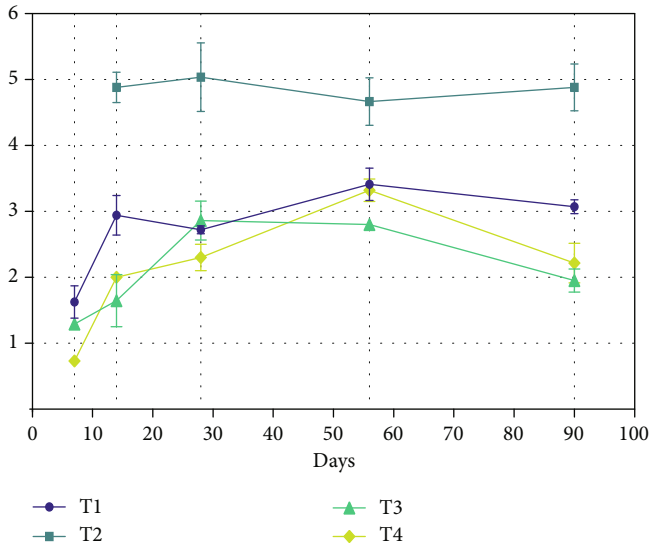

Figure 12: Lactic-to-acetic ratio; kinetics by experimental treatment ($p < 0.001$) over a conservation period of 90 days. Conservation time effect ($p < 0.001$) assessed at 7, 14, 28, 56, and 90 days. Interaction of treatment × conservation time ($p = 0.087$). T0 not assessed.

and T2 (Table 4) and, that common NaCl is a source of chlorine and sodium, we could speculate that the addition of these ions to the silage could have an effect on the CEC activity, reducing the BC in treatment T2.

However, less concentration of organic acids detected in T2 did not have a significant effect on either fermentative losses (except for a small increase in GLs as previously mentioned; Table 4) or pH, which could be explained since the lower BC reported in this treatment allows an adequate drop in pH, despite its lower (but not statistically significant) concentrations of lactic acid when compared with treatment T1.

NaCl addition in treatment T2 also resulted in a higher initial SC concentration compared to treatment T1 (Table 4). This could be influenced by the nature of refractometry as an indirect method to determine the SC concentrations in this study. Indirect measurement of soluble solids by refractometry is a quick and inexpensive method widely used to determine sugar content in sugar cane, sugar beet, and table beet, among other plants and vegetables [58]. However, it measures the total soluble solids, which include sugars, but also other diluted substances like organic acids, minerals, and amino acids [59]. Therefore, we could speculate that added NaCl as a mineral source and soluble solid could lead to a bias, increasing the specific refractive index with respect to the concentration of solutes in treatment without NaCl addition.

A reduction in NH_3-N concentrations (7.82% and 9.56% of total N) and BC (44.2 and 14.2 mE NaOH/100 g DM) was observed in treatments with forage addition when compared to T1 and T2 treatments (Figures 8 and 9, respectively). Conversely, the highest significant values of mass T°C and pH values, accompanied by lower lactic acid concentration after the 90-day conservation period, were observed in silages with forage (T3 and T4). Higher moisture and SC contents in silages could increase proteolytic activity, which generally results in higher soluble N and NH_3-N concentrations compared to drier silages [48, 60]. This could explain the reported results of this study where a lower NH_3-N concentration was found in drier (T3 and T4) compared to wet silages without forage addition (T1 and T2; Table 4 and Figure 8). Additionally, higher moisture silages could lead to microbial safety concerns since this condition is highly associated with undesirable clostridial fermentations [22]. Higher than normal acetic acid concentration and pH

values, as well as high concentrations of NH_3-N or propionic acid (> 0.3%–0.5%) with contrasting lower than normal lactic acid concentrations, are some of the parameters that indicate possible clostridial fermentation activity in the ensiled mass [36]. Nevertheless, none of these situations were detected in the wet silages of this study, as discussed throughout this section.

Despite the lowest fermentative losses reported by T3, its lower moisture and SC contents could also explain the lower lactic and acetic acid concentrations, leading to a higher (although technically acceptable) pH value, compared to T1 (Table 4 and Figure 7). These parameters are strongly associated with organic acid synthesis, responsible for defining a proper silage pH. According to Kung Jr. et al. [36], the decrease in silage pH depends on appropriate lactic acid concentrations, which is a 10–12 times stronger acid (pKa of 3.86) compared to acetic acid (pKa of 4.75). Acetic acid, on the other hand, has more attributes in terms of greater aerobic silage stability, based on its ability to control fungal growth. Regarding propionic acid concentrations, these are typically found to be below 0.1% or undetectable in good-quality silages. In this study, propionic acid concentrations were undetectable by HPLC analysis in all the assessed treatments (Table 4).

Furthermore, pH is the main parameter affected by BC, since this refers to the resistance of the ensiled mass to the progressive and accelerated decrease in pH [22, 45]. Contrary to expectations, treatments with the highest BC were those with the lowest final pH (Table 4; T1 and T2) and the best kinetics in terms of the drop of pH over time, which stabilized after 28 conservation days. Treatment T3 showed a slow drop in pH until 28 conservation days, after which it showed a turning point reaching its highest value at 90 conservation days (Figure 7). It is still unclear why pH showed an inverse performance in relation to BC in all treatments.

Cherney and Cherney [35] suggested that silages made from mature grass are poor in CEC and, consequently, in buffer capacity. Although those silages ferment more slowly, they could finally have a greater buffering effect than expected. This might allow us to speculate that a higher content of moisture and SC on wet silages (as T1 and T2 reporting 21% of DM and 12% of SC) and a proper content of moisture and SC in T3 (68.5% and 7.75%, respectively), which promotes higher activity of LAB with a derivative increase in lactic acid concentrations, are much more decisive in defining its pH values compared to BC effect, as reported in this study.

Related to T4, it was the treatment with the highest internal T°C throughout all conservation periods evaluated (Figure 2), followed by T3 and finally T1 and T2, which did not show significant differences in T°C after 90 conservation days. T°C of ensiled mass can affect microbial activity, fermentation products and nutritive value of silage, being a 20°C–30°C range desirable for silage fermentation and optimal conservation [61]. Higher T°C reported in T4 could be associated with fungal activity which deteriorated the silage mass, with detrimental effects on quality parameters (Table 4). This finding contrasts with studies reporting as necessary a T°C increase by approximately 10°C for incurring in a substantial DM loss [35]. Although this study demonstrated significant variations in T°C due to the effect of treatment and time (Figure 5), it can be concluded that such variations are not substantial in practice, as all silages exhibited average T°Cs within the acceptable threshold for optimal fermentation, as previously mentioned.

3.4. D Assessing. The calculation of the *D* showed that T3 was the treatment with the fermentation products and conservation characteristics closest to the ideal in the period between 28 and 56 days, surpassed only by T1 when the conservation period was extended to 90 days (Table 5). Considering that the efficiency of the conservation process is judged not only by the quality of the final product but also by the length of time the ensiled substrate can be preserved, the results presented for periods prior to the final conservation time are less relevant in determining the most effective treatment. However, they do give an idea of the progress and changes over time.

Although the highest *D* on the scale is 1, the best treatment was T3 after 28 conservation days ($D = 0.5$), whereas the best after the 90 days period was T1 ($D = 0.365$), closely followed by T2 ($D = 0.355$) and, with the lower index, T3 ($D = 0.253$).

On the other hand, the reference parameters considered as "ideal" correspond to the final fermentation products and conservation characteristics of corn silage with initial DM around 30-40% after a period which usually ranges between 90 and 120 days. However, it could not be entirely appropriate to weight the *D* for the previous phases (*D* of silages at opening times from 7 to 56 days), taking as a reference the products and characteristics expected at the end of the process.

Another important aspect to take into account when interpreting the results of the *D* approach is that fruit silages rich in moisture and SC (T1 and T2) and another with high DM content close to 60% (T4) were compared with a reference parameter that, by default, is much closer to the initial characteristics of the treatment with forage inclusion and initial DM content of 31.6% (T3). This could have influenced the highest desirability indices between 28 and 56 conservation days in this treatment, surpassed only by T1 and T2 after 90 days, due to its higher final SC concentrations, its lower and more desirable pH and a trend to higher and more desirable lactic acid concentrations. It should be noteworthy that fermentation and ensilability characteristics of corn silage with initial DM around 30%–40% were considered as reference, as this silage has historically shown the most desirable fermentation and ensilability performances. Furthermore, contrary to legume silages with high or low moisture contents, or grass and wet corn grain silages, there are no agreed reference values in the literature for fruit silages with high moisture and SC content as it was our case in the present study. This fact underlines the necessity for further research to provide a clearer and more reliable guide for performing future studies assessing fruit silage and its full potential for feedstuffs conservation as a sustainable alternative for planning adequate livestock feeding and nutrition schedules in the tropics.

TABLE 5: Desirability index values per treatment in each conservation time (from 7 to 90 days).

Treatment	Conservation time				
	7 days	14 days	28 days	56 days	90 days
T0	0.262$^{4°}$	0.183$^{5°}$	0.232$^{4°}$	0.090$^{5°}$	0.090$^{5°}$
T1	0.325$^{2°}$	**0.478** $^{1°}$	0.450$^{2°}$	0.376$^{3°}$	**0.365** $^{1°}$
T2	**0.332** $^{1°}$	0.314$^{2°}$	0.402$^{3°}$	0.401$^{2°}$	0.355$^{2°}$
T3	0.246$^{5°}$	0.258$^{3°}$	**0.500** $^{1°}$	**0.434** $^{1°}$	0.253$^{3°}$
T4	0.281$^{3°}$	0.205$^{4°}$	0.198$^{5°}$	0.097$^{4°}$	0.097$^{4°}$

Note: Superscripted ordinal number: Ranking position of treatment into each assessed conservation time. Higher desirability index (*D*) by conservation time is in bold.

Abbreviations: **T0**: ripe calabash tree fruit (unground and not ensiled); **T1**: ground fruit without any additional additives; **T2**: ground fruit +1.5% of salt (saline silage); **T3**: mixture of ground fruits and foliage of calabash tree in a 30:70 fruit-to-roughage ratio; **T4**: mixture of ground calabash tree fruits and angleton hay in a 50:50 fruit-to-roughage ratio.

4. Conclusions

The hard shell of the CTF is insufficient for preserving the pulp and its nutritional value for more than 28 days.

The significant increase in DM and the fibrous fraction in the T4 treatment mixture, due to the high proportion of Angleton hay, resulted in physical and bromatological changes to the silage. This increased losses and significantly altered the fermentation profile, reducing its conservation potential and nutritive value.

Adding NaCl at a rate of 1.5% of the ensiled mass (T2) did not improve the fermentation or conservation parameters of CTF silage. However, it did increase GLs and reduce the concentration of organic acids, though it had no effect on the pH. It can be concluded that the use of NaCl as a preservative for CTF is unnecessary.

This study has demonstrated the feasibility of the silage technique as a method for preserving CTF and suggests fruit silage without any additives followed by silage of ground fruit and calabash tree foliage mixture (30:70 fruit-to-roughage ratio) as efficient alternatives for preserving the nutritional quality of these feeds to supplement ruminant diets during dry season shortages in tropical farming systems. However, mixing CTF with tropical forages and other substrates may be a viable option if (i) the DM and NDF contents do not exceed 40% and 45%, respectively, and (ii) adding forage to the mixture significantly improves the chemical composition and nutritive value of the silage, without compromising its fermentative profile or the way it is conserved.

Notwithstanding the encouraging results of this study, its limitations must be acknowledged. The novelty and the use of experimental minisilos as the study's framework could hinder its scalability due to the unavoidable changes derived from environmental differences and demand for additional labor during the preparation of large-scale silages under farm conditions. Consequently, further research is necessary to evaluate the reproducibility and scalability of these results.

Nomenclature

ADF acid detergent fiber
ANOVA analysis of variance
AOAC Association of Official Analytical Chemists
BC buffering capacity
CP crude protein
CTF calabash tree fruit
DM dry matter
DMR dry matter recovery
EE ether extract
FC fibrous carbohydrates
FM fresh matter
FML fresh matter losses
GE gross energy
GL gas losses
LAB lactic acid bacteria
NDF neutral detergent fiber
NFC nonfibrous carbohydrates
NH_3-N ammoniacal nitrogen
OM organic matter
SC soluble carbohydrates
TC total carbohydrates

Data Availability Statement

The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available due to privacy or ethical restrictions.

Conflicts of Interest

The authors declare no conflicts of interest.

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Supporting Information

Additional supporting information can be found online in the Supporting Information section.. *(Supporting Information)* Figure S1: The unripe fruit of the calabash tree (*Crescentia cujete*) is left on the tree (not harvested). Figure S2: Ripe fruit of the calabash tree (*Crescentia cujete*) hanging from the tree (not harvested). Figure S3: The pulp content of ripe (4 months old) fruit of the calabash tree (*Crescentia cujete*) is shown after harvesting and opening. Figure S4: Visual aspect of calabash tree fruits in treatment T0 (not ensiled fruits) after a 28-day conservation period. Figure S5: Visual aspect of calabash tree fruits in treatment T0 (not ensiled fruits) after a 56-day conservation period. Figure S6: Visual aspect of calabash tree fruits in treatment T0 (not ensiled fruits) after a 90-day conservation period, in which there was a high presence of fungus inside all the opened fruits. Figure S7: Visual aspect of calabash tree fruits in treatment T0 (not ensiled fruits) after a 90-day conservation period in the presence of unidentified living organisms parasitizing the external surface of the peel. Figure S8: Visual aspect of the calabash tree fruits in treatment T0 (not ensiled fruits) after a 90-day conservation period showing a severe reduction in pulp content and a high fungal presence. Figure S9: Visual aspect of calabash tree fruits in treatment T0 (not ensiled fruits) after a 90-day conservation period in the presence of larvae inside the fruits. Figure S10: High presence of fungus in the experimental mini-silos after a 28-day conservation period in treatment T4 (calabash tree fruit and Angleton hay silage at a 50:50 fruit-to-hay ratio). Figure S11: High presence of fungus in the experimental mini-silos after a 56-day conservation period in treatment T4 (calabash tree fruit and Angleton hay silage at a 50:50 fruit-to-hay ratio). Figure S12: High presence of fungus in the experimental mini-silos after a 90-day conservation period in treatment T4 (calabash tree fruit and Angleton hay silage at a 50:50 fruit-to-hay ratio).

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