Fermentation Profile, Nutritional Value, and Microbial Population of C4 Grasses Silages with or Without Bacterial Additive

Sarah E. E. Bernardo
Universidade Estadual do Norte Fluminense: Universidade Estadual do Norte Fluminense Darcy Ribeiro
Paulo H. B. Chrisostomo
Universidade Federal Rural do Rio de Janeiro: Universidade Federal Rural do Rio de Janeiro
Michele G. Camilo
Universidade Estadual do Norte Fluminense: Universidade Estadual do Norte Fluminense Darcy Ribeiro
Danielle F. Baffa
Universidade Estadual do Norte Fluminense: Universidade Estadual do Norte Fluminense Darcy Ribeiro
Elizabeth F. Processi
Universidade Federal Rural do Rio de Janeiro: Universidade Federal Rural do Rio de Janeiro
Alberto M. Fernandes
Universidade Estadual do Norte Fluminense: Universidade Estadual do Norte Fluminense Darcy Ribeiro
Tadeu Silva de Oliveira (tsoliveira@uenf.br)
Universidade Estadual do Norte Fluminense Darcy Ribeiro https://orcid.org/0000-0001-7703-9323

Research Article

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Fermentation profile, nutritional value, and microbial population of C4 grasses silages with or without bacterial additive

Sarah E. E. Bernardo[1]. Paulo H. B. Chrisostomo[2]. Michele G. Camilo[1]. Danielle F. Baffa[1]. Elizabeth F. Processi[2]. Alberto M. Fernandes[1]. Tadeu S. Oliveira[1]

Abstract - The objective of the present study was to evaluate the use of bacterial additive (*Lactobacillus plantarum* and *Propionibacterium acidipropionici*) on chemical composition, *in vitro* gas production, pH, losses, aerobic stability, and microbial population of corn, pearl millet, and sorghum silages in plastic bags silos (without *vacuum*). The experiment was carried out in a randomized block design, in a 2 × 3 factorial scheme, with or without additive ([Control] without additive and *Lactobacillus plantarum* [2.5 × 1010 cfu/g] and *Propionibacterium acidipropionici* [2.5 × 1010 cfu/g] Biomax corn, Lallemand, Saint-Simon, France [LP]) and three crops of agricultural interest; pearl millet, sorghum, and corn, with four replicates per treatment. We performed chemical analyses and *in vitro* gas production to determine the nutritional value of the silages. We also evaluated the aerobic stability, ammoniacal nitrogen (NH_3), pH, and microbial population of the silages. The additive increased the crude protein content (P = 0.0062) in corn and sorghum and decreased the LIG content (P = 0.0567). The gas production was not affected (P >0.05) by the additive and neither between crops. In aerobic stability, we observed that the additive affected the temperature of the sorghum silage (P = 0.0123). The additive decreased NH_3 (P = 0.0095) content. The additive increased (P = 0.0441) the lactic acid bacteria population in the pearl millet, corn, and sorghum silages. Thus, the bacterial additive did not improve the fermentation profile and nutritional value of corn, pearl millet, and sorghum silages in plastic bag silos.

Keywords: Conservation. Fermentation capacity. Inoculant. Lactic acid bacteria

Introduction

In Brazil, milk and beef productions are based on pastures, as they are a less expensive feed source for the farmers. However, the seasonality of forage production is a critical

Tadeu S. de Oliveira
tsoliveira@uenf.br
[1]Laboratory of Animal Science, State University of Northern of Rio de Janeiro, Campos dos Goytacazes, RJ 28013-602, Brazil.
[2]Federal Rural University of Rio de Janeiro, Campos dos Goytacazes, 28022-560, Brazil.

point for planning livestock activity. Under such conditions, it is necessary to establish feeding strategies for the herds in which the production and conservation of supplementary forage must be considered (Chaudhry, 2008). The most widespread methods for preserving feed are haymaking and ensiling.

The storage of forage through the ensiling technique receives a greater emphasis on the part of the farmers for presenting excellent results. Still, some factors must be considered, and the main one is respecting the characteristics of each crop, as they can directly influence the silage quality. Specifically for the ensiling, the final quality of the feed is directly related to the material that gave rise to it and the conditions in which it was ensiled (Jobim et al., 2007).

During the ensiling process, another critical factor is the additive. The microbial inoculants are recommended to reduce losses in silage of tropical grasses since homolactic bacteria compete with epiphytic microorganisms, increasing the fermentation efficiency (Kung Jr et al., 2003; Borreani et al., 2018). Inoculants are one of the main additives in the ensiling, aiming to dominate fermentation through the rapid production of lactic acid (homolactic bacteria) and consequent pH decrease, inhibiting the growth of undesirable microorganisms and production of acetic and propionic acid (Kung Jr. et al., 2003). Other bacteria, such as heterofermentative, can increase acetic and propionic acid (Kung Jr. et al., 2003; Zopollatto et al., 2009; Bernardes and Rêgo, 2014). There are several compositions of inoculants on the market. As a rule, those produced from homolactic bacteria improve the fermentation pattern, whereas heterolactic bacteria inoculants are used to increase aerobic stability (Queiroz et al., 2018). *Lactobacillus plantarum* is one of the most used among homolactic bacteria due to its vigorous growth, acid tolerance, and a high potential for lactic acid production (Muck, 2010). In the heterofermentative bacteria group, the *Propionibacterium acidipropionici* uses lactic acid

and glucose as substrates to produce acetic and propionic acid, which effectively control fungi under low pH (Zopollatto et al., 2009).

Therefore, we hypothesized that the bacterial additive would not affect the fermentation of C4 grasses silages. Thus, the objective of the present study was to evaluate the bacterial additive (*Lactobacillus plantarum* and *Propionibacterium acidipropionici*) on chemical composition, in vitro gas production, pH, losses, aerobic stability, and microbial population of corn, pearl millet, and sorghum silages in plastic bag silos (without *vacuum*).

Material and Methods

Location

The experiment was carried out at the Universidade Federal Rural do Rio de Janeiro (UFRRJ) - Campos dos Goytacazes *Campus*, RJ, Brazil (21°47'54"S and 41°17'34"W, 12 m a.s.l.) and at the Animal Science Laboratory of the Universidade Estadual do Norte Fluminense Darcy Ribeiro (UENF) - Campos dos Goytacazes, RJ, Brazil (21°45'41"S and 41°17'27"W, 10 m a.s.l.). According to Köppen's classification (Alvares et al., 2013), the climate in the North of the Rio de Janeiro State is classified as Aw, i.e., humid tropical with a rainy summer, dry winter, and annual rainfall around 1,020 mm. The soil of the experimental area is classified as Cambisol.

Ensiling, additive, and experimental design

The pearl millet, corn, and sorghum were harvested and processed in a stationary forage chopper (JF Maxxium Model, JF Máquinas Agrícolas LTDA, Brazil) with an average particle size of ± 1.5 cm. We used plastic bags (polyethylene 51 cm wide × 110 cm long and 200 microns) for ensiling the forages. The silos were closed with nylon clamps and stored at

an ambient temperature of 25±2.3 ºC for 90 days. The silos were packed with a density of 600 kg/m3 (fresh forage ensiled).

The experiment was carried out in randomized blocks in a 2 × 3 factorial scheme, with or without the additive ([Control, CON] without additive and *Lactobacillus plantarum* [2.5 × 1010 cfu/g] and *Propionibacterium acidipropionici* [2.5 × 1010 cfu/g] Biomax corn, Lallemand, Saint-Simon, France [LP]) and three crops of agricultural interest: pearl millet (*Pennisetum glaucum* L.), cv. BRS 1501; sorghum (*Sorghum bicolor* x *Sorghum sudanense*), hybrid BRS810; and corn (*Zea Mays* L.), cv. PR1150. We used four replicates per treatment, totaling 48 experimental units. The treatments were randomly distributed in the silos. The microbial inoculant was used according to the manufacturer's recommendations (2 g/ton of forage), it was diluted in water and sprayed on the forages to be ensiled.

Chemical composition

After opening the silos, the silage samples were dried in a forced-air oven at ±55 °C for 72 hours to yield the partially dried samples. Then, the samples were ground in a Wiley mill fitted with a 1-mm-sieve. We determined the contents of total dry matter (DM, AOAC Method 967.03, AOAC, 1990), crude fat (CF, AOAC Method 2003.06; Thiex et al., 2003), ash (ASH, AOAC Method 942.05, AOAC, 1990), crude protein ([N×6.25]CP, AOAC Method 984.13 and AOAC Method 2001.11; Thiex et al., 2002), neutral detergent fiber using a standardized heat stable amylase solution and the expressed results with residual ash (aNDF, AOAC Method 2002.04; Mertens, 2002) and lignin (LIG) (Möller, 2009). The non-fibrous carbohydrate (NFC) content was estimated as: $NFC(g/kg) = 1000 - CP - CF - Ash - NDF$.

The hemicellulose was calculated by the difference between the NDF and ADF contents, and the cellulose by the difference between ADF and Lignin contents, expressed in g/kg

DM.

Gas production kinetics

The Ethics Committee on the Use of Experimental Animals approved all experimental procedures, 419 protocol. We collected the ruminal fluid from three sheep, 45 kg (standard deviation = 3.2 kg), with permanent rumen cannulas. The animals were housed in collective pens with troughs and drinkers. The sheep were adapted to a diet with Tifton 85 hay and concentrate feed for 14 days to meet the maintenance requirements. After this period, the collection of ruminal fluid began, and it took place moments before daytime feeding, as recommended by Yáñez-Ruiz et al. (2016). The ruminal fluid (liquid and solid) was collected at several points of the liquid-solid interface of the ruminal environment for each incubation battery. We used about 500 mg (standard deviation = 10 mg) of silage sample in amber penicillin flasks together with 50 ml of an inoculum (1:4 ratio, ruminal fluid, and buffer solution, respectively). The buffer solution was prepared as described by McDougall (1948). The flasks were immediately filled with CO_2, closed, and placed in a water bath.

The time profiles of gas production were obtained using a non-automated device, similar to Abreu et al. (2014). We measured pressure and volume at times 0; 1; 2; 3; 4; 6; 8; 10;12; 16; 20; 24; 30; 36; 48; 72 and 96 hours after the addition of ruminal inoculum. The cumulative pressure and volume of fermentation gases were obtained by summing the corrected readings throughout the measurement times.

We used the model described by Groot et al. (1996) to explain the cumulative gas production profiles:

$$G = A/(1 + (B^c/t^c)) \quad \text{Eq.1}$$

$$R_M(mLh^{-1}) = B \times (C-1)^{1/C} \quad \text{Eq.2}$$

Where, the parameter G represents the amount of gas produced per unit of organic matter incubated at time *t* after the incubation period; the parameter A represents the asymptotic gas

production (mg/g OM); the parameter B is the time (h) after incubation in which half of the asymptotic gas was produced, it represents the gas production speed; the parameter C is a constant that determines the sharpness of the curve change characteristic. The R_M represents the maximum gas production rate when the microbial population does not limit the fermentation and digestion is not reduced by chemical or structural barriers of the potentially digestible matter.

Aerobic stability test

About 2.0 kg of silage was packed in plastic bags of 5.0 kg capacity, where it remained for seven days in a room with a controlled temperature (25°C) to assess aerobic stability. For this, we used a data logger (Log 110 EXF Inconterm; Brazil) inserted in the ensiled mass at a depth of 10 cm in the central portion, and the temperature recording was carried out at intervals of 8 hours. Aerobic stability was calculated as the time, in hours, in which the silages had a temperature 2 °C higher than the ambient temperature after opening the silo. In addition, we collected samples from each silo during the aerobic stability assessment at 24-hour intervals to determine DM (AOAC Method 967.03, AOAC, 1990) (Kung Jr. et al., 2003).

Fermentative parameters

After opening each silo, the material was homogenized, a sample of 25 g of fresh silage was taken, and 225 ml of saline solution (8.5 g of NaCl/L of distilled water) was added and homogenized for 1 minute in an industrial processor. The extract was filtered through a double layer of gauze, and pH was measured with a pH meter (MPA-210, Tecnopon, Brazil) (Kung Jr., 1996). Aliquots of 2 mL of extract were transferred to test tubes with 1 mL of

157 sulfuric acid (1N) and stored at −20◦C. Ammoniacal nitrogen analysis was performed
158 according to the methodology of Fenner (1965).
159
160 **Microbial population**
161 A 10 ml aliquot of the aqueous extract was submitted to serial dilutions (10-1 to 10-6). The
162 cultivation of microorganisms was performed in sterile Petri dishes. We used the VRB (Violet
163 Red Bile) culture media to count enterobacteria with an incubation period of 24 h at 37 °C;
164 for the fungi counting, we used the PDA (Potato Dextrose Ágar) culture media with an
165 incubation period of four days at 25 °C; and we used the MRS (De Man, Rogosa, Sharpe)
166 culture media, for 48 h at 37 °C, to count lactic acid bacteria. We counted the dishes that
167 showed between 30 and 300 colony-forming units (CFU). The results were transformed into a
168 logarithmic basis (log10 cfu) for the data evaluation and interpretation.
169
170 **Statistical Analysis**
171 Data regarding chemical composition, losses, ammoniacal nitrogen, pH, temperature,
172 microbial population, and cumulative gas production were analyzed in randomized blocks in a
173 3 × 2 factorial scheme with four replicates. The data were compared through the Tukey test
174 with a 0.05 significance level using the SAS MIXED package (SAS University Edition, SAS
175 Institute Inc., Cary, NC, USA).
176 We used the following statistical model:
177 $$Y_{ijk} = \mu + \alpha_i + \beta_j + \alpha\beta_{ij} + b_k + e_{ijk}$$
178 Where: Y_{ijk} is the value observed for the variable under study referring to the *k-th* replicate of
179 the combination of the *i-th* level of factor α with the *j-th* level of factor β; μ is the mean of all
180 experimental units for the variable under study; α_i is the use or not of additive in the silage
181 with i = 1.2; β_j is the crop effect, with j = 1,2,3; $\alpha\beta_{ij}$ is the interaction between the use or not

of additive and crops; b_k is the effect of the *k-th* block on the observation; and e_{ijk} is the error associated with observation Y_{ijkl}.

The pH and temperature data (aerobic stability) were analyzed as repeated measures over time, and we applied regression analysis with a significance level of 0.05 using the SAS MIXED package (SAS University Edition, SAS Institute Inc., Cary, NC, USA).

We used the following statistical model:

$$Y_{ijkl} = \mu + \alpha_i + \beta_j + \tau_k + \alpha\beta_{ij} + \alpha\tau_{ik} + \beta\tau_{jk} + \alpha\beta\tau_{ijk} + b_l + e_{ijkl}$$

Where: Y_{ijkl} is the value observed for the variable under study referring to the *l-th* replicate of the combination of the *i-th* level of factor α with the *j-th* level of factor β in the *k-th* hour; μ is the mean of all experimental units for the variable under study; α_i is the use or not of additive in the silage with i = 1.2; β_j is the crop effect, with j = 1,2,3; τ_k is the random effect of the evaluation hours with k = 0.24,...,144 for pH and 0.8,16,...,162 for temperature; $\alpha\beta_{ij}$is the interaction between the use or not of additive and crops; $\alpha\tau_{ik}$ is the interaction between the use of additive or not and the evaluation hours; $\beta\tau_{jk}$ is the interaction between crops and evaluation hours; $\alpha\beta\tau_{ijk}$ is the interaction between the use of additive or not, crops, and evaluation hours; b_k is the effect of the *l-th* block on the observation; and e_{ijkl} is the error associated with observation Y_{ijkl}.

Results

Chemical composition and gas production

There was no interaction effect (P >0.05) between additive and crops (Tables 1), except for CP content (P = 0.0357). The use of additive increased the CP content (P = 0.0062) and reduced the LIG content (P = 0.0567) in corn (19.65% [42.06/52.35]) and sorghum (14.43% [50.15/58.61]) silages. The CF (P = 0.7695), NDF (P = 0.0607), and NCF (P = 0.1429) contents did not show any statistical difference (Tables 1 and 2) between crops.

The gas production kinetics was not affected (P >0.05) by the additive and neither between crops (Tables 1 and 3).

Aerobic stability test

There was no interaction effect (P >0.05) between additive and crops (Tables 1) and additive and time for aerobic stability (Figure 2). Regarding the aerobic stability, we observed that the additive did not affect the temperature of the corn (P = 0.6419) and pearl millet (P = 0.8527) silages over the days. However, the additive affected (P = 0.0123) the temperature of the sorghum silage (Figure 2C). The additive only affected the pH of the pearl millet silage (P = 0.0065) (Figure 2D).

Fermentation parameters

There was no interaction effect (P >0.05) between additive and crops (Tables 1). The temperature right after the opening of the silos and dry matter losses were not affected (P > 0.05) by the additive (P >0.05) or between crops (P >0.05) (Tables 1 and 4). The additive decreased by 1.09% the pH of corn silage, 10.67% of sorghum silage, and 3.37% of pearl millet silage. There was a statistical difference (P <0.0001) between crops (Tables 1 and 4).

Ammoniacal nitrogen followed a similar behavior to pH. The additive increased 44.15% of the ammonia nitrogen content in corn silage, 25.91% in sorghum silage, and decreased about 5% in pearl millet silage. The crops differed significantly (P = 0.0095) (Tables 1 and 4).

Microbial population

There was no interaction effect (P >0.05) between additive and crops (Tables 1). There was no appearance of enterobacteria in the silages with or without the additive. The additive increased (P = 0.0441) by 9.63%, 19.46%, and 31.27% the population of lactic acid bacteria

232 in the pearl millet, corn, and sorghum silages, respectively (Table 5). Corn silage presented a
233 more significant amount of fungi (P <0.0001) than pearl millet and sorghum silages,
234 regardless of the use of additive (Table 5).
235
236 **Discussion**
237 The nutritional value of silage depends mainly on three factors: the genetic material (plant
238 biomass, grain production, drought tolerance, and disease resistance), the stage at which the
239 forage was harvested (it affects the composition and the final quality of the preserved forage,
240 and the stage indicated for ensiling is between the doughy and farinaceous stages) and the
241 microorganisms (epiphytic) present in the forage during the ensiling process (Oliveira et al.,
242 2011; Behling Neto et al., 2017). Thus, we observed the chemical composition of the silages
243 and noticed a difference between the crops, except for the CF, NDF, and NCF contents (Table
244 2). The lower DM content observed in pearl millet and sorghum silages can be explained by
245 the higher resistance to drought due to an adaptive process that prevents excessive
246 dehydration such as smaller stomata, early stomatal closure, low stomatal density, and
247 increased leaf serosity, retaining more water in the plant (Levitt, 1980). In addition to the
248 difference between crops, we also observed that the additive increased the CP contents (Table
249 2), but the additive could not reduce losses caused by proteolysis. Analyzing the ammoniacal
250 nitrogen (Table 4), we observed that the CP degradation was between 25 and 30%. For Kung
251 Jr et al. (2018), the rapid pH decline is crucial to reduce protein degradation during ensiling,
252 which probably happened in corn silage without additive, as it had a low degradation rate
253 (21.11% [8.88/42.06]). Another factor that may have influenced the protein degradation is the
254 material of the silo (polyethylene) in terms of oxygen permeability, i.e., gas exchanges
255 between the silage and the external environment, with oxygen entering even without the
256 plastic bags present any physical damage (Amaral et al., 2014) which may have allowed the

increase of undesirable microorganisms such as mold (Table 5). These microorganisms have their activity intensified in the presence of soluble carbohydrates, acids, and proteins, increasing the silage pH (Table 4). The hemicellulose degradation has been neglected for many years (Ning et al., 2017), but some studies have shown this degradation occurs during the ensiling (Muck, 1990; Chen et al., 2015). Melvin (1965) and Yahaya et al. (2001) reported that the degradation products of hemicellulose (xylose) and starch (glucose) could be substrates for microorganisms to produce acids during the ensiling; this fact was observed in our study (Table 2). LIG and CEL contents were lower in corn silage (Table 2). These values can be explained by the carbon translocation from the leaf to the formation and filling of grains, causing an increase in starch contents (Di Fonzo et al., 1982). In our study, the NFC contents were not different (P = 0.1429) between crops, but corn silage was 2.35% higher than pearl millet silage and 11.36% higher than sorghum silage without additive (Table 2). Corn silage presented the lowest ash content (Table 2). According to Ashbell (1995), the low ash content indicates excellent forage conservation, as the occurrence of inadequate fermentation results in losses of organic matter, increasing the share of ash in DM.

The *in vitro* gas production allows evaluating the kinetics of ruminal fermentation, providing information on the rate and extent of feed degradation in the rumen (Theodorou et al., 1994). In our study, the gas production was not affected (P > 0.05) by the additive and neither between crops (Table 1 and Figure 1). For Bach et al. (2005), crude protein concentrations below 70 g/kg may restrict microbial activity due to lack of nitrogen. In this study (Table 2), these concentrations were from 40 to 60 g/kg. However, we observed that although there was no difference (P > 0.05), the time taken for half of the asymptotic gas to be produced (Parameter B) in the corn silage with or without additive was shorter than the other silages (Table 3). Gas production rates peaked in the first hours of incubation, being

281 longer in the corn silage without additive (Figure 1C), but all silages had a final rate below
282 0.1 ml/h (Figure 1).

283 The aerobic stability of the silage is expressed as the resistance of forage mass to
284 deterioration after opening the silo, i.e., the speed at which the mass deteriorates after its
285 contact with the air (Jobim et al., 2007). Thus, we observed that the silages' temperature at the
286 opening was not affected by the additive (P = 0.8911) and neither between crops (P =
287 0.3196). However, pH was affected by the additive (P = 0.0013) and crops (P <0.0001)
288 (Table 4). *Lactobacillus plantarum*, one of the inoculants in this study, aims to increase lactic
289 acid production, consequently reducing the pH of the ensiled mass and inhibiting the growth
290 of unwanted microorganisms (McDonald et al., 1991; Muck, 2010). Analyzing the
291 temperature over the days, we observed that in the first 36 hours, there was a peak
292 temperature in sorghum silage regardless of the additive. However, the silage with an additive
293 reduced its temperature more quickly (Figure 2C). For Woolford (1990), the initial increase in
294 temperature is caused by the growth of yeasts and filamentous fungi, but after some time,
295 according to Muck and Pitt (1992), the increase in pH (above 5.0) can favor the growth of
296 bacilli that can cause a second increase in the temperature, this fact was observed in our study
297 (Figures 2 B and C). The plastic bag (polyethylene) silos can present oxygen permeability at a
298 temperature of 25 °C. Gas exchange between the interior of the silo and the environment is
299 close to 1 liter/m2, which is a value for an intact bag without any physical damage (Greenhill,
300 1964). This exchange can make the silage more prone to aerobic deterioration due to the
301 increase in the permeability of the bags, as the aeration of this mass allows the action of
302 yeasts that oxidize the silage's preservative organic acids triggering aerobic degradation and
303 increasing pH. In our study, the pearl millet, corn, and sorghum silages without additive
304 increased 2.0 points in pH in 24, 96, and 96 hours, respectively, whereas those with additive,

increased 2.0 points in pH in 48 hours (Figures 2D, E, and F). The aerobic stability loss of silages is usually manifested by an increase in temperature and a change in pH.

Ammoniacal nitrogen (NH_3-N) indicates the amount of protein degraded during the fermentation. It is an indicator of the extent of clostridial activity since it is produced in small amounts by other microorganisms in the silage and plant enzymes (Borreani et al., 2018). According to Blajman et al. (2020), the combination of hetero and homofermentative bacteria favors reducing pH values, ammoniacal nitrogen, and fermentative losses in silages. The NH3-N levels were affected by the additive (0.0040) and between crops (P = 0.0095) (Table 4). According to Tomich et al. (2003), NH_3-N values below 10 g/kg CP indicate good fermentation, and above 15 g/kg CP of NH_3-N in silage indicate a significant amount of proteolysis. In the present study, only corn silage without additive (8.88 g/kg CP) presented NH_3-N values below 10 g/kg CP. This fact may indicate a higher intensity of proteolysis, especially by amino acid degradation by bacteria of the genus *Clostridium* (Diether and Willing, 2019).

It is also essential to understand the microbial population, as the ensiling will preserve the forage and inhibit undesired microorganisms (*Clostridium* sp, enterobacteria, yeasts, and fungi), influencing the silage quality (Muck, 2010). In this study, we observed there were no counts for enterobacteria in the silos. This fact is related to the active growth of lactic acid bacteria (LAB) during the fermentation process, as the pH decrease to values between 3.35 and 3.75 (Table 4), it favors the population decline of enterobacteria rapidly, turning LAB the main microorganisms in silage (McDonald et al., 1991). The additive increased the LAB population and decreased the pH of the silos. *Lactobacillus plantarum* (one of the microorganisms in the additive of this study) aims to increase the lactic acid production, consequently reducing the pH of the ensiled mass and inhibiting the growth of unwanted microorganisms (McDonald et al., 1991; Muck, 2010). Analyzing the crops, we observed that

corn silage presented the highest (P <0.001) fungi population, about 37.75% more than pearl millet silage and 29.27% more than sorghum silage (Table 5). This fact is probably because the plastic bag (polyethylene) silos can present oxygen permeability, and most fungi are strictly aerobic. They can use sugars (glucose, sucrose, and maltose) and more complex compounds (starch, cellulose, and hemicellulose) as substrates for their growth (Wang et al., 2020).

Conclusion

The bacterial additive did not improve the fermentation profile and nutritional value of corn, pearl millet, and sorghum silages in plastic bag silos.

Declarations

Authors' contributions

Conceptualization: A. M. Fernandes and T.S. Oliveira. Data curation: T.S. Oliveira. Formal analysis: T.S. Oliveira. Investigation: E.F. Processi, and T.S. Oliveira. Methodology: S.E.E. Bernardo, E.F. Processi, M.G. Camilo, D.F. Baffa, and P.H.B. Chrisostomo. Resources: T.S. Oliveira. Supervision: T.S. Oliveira. Writing-original draft: T.S. Oliveira. Writing-review & editing: A. M Fernandes, S.E.E. Bernardo, and T.S. Oliveira.

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Consent to participate: Not applicable.

Consent for publication: Not applicable.

Availability of data and material: Not applicable.

Code availability: Not applicable.

Research involving human participants and/or animals

Ethical Approval: All experimental procedures were approved by the Ethics Committee on the Use of Experimental Animals of the Universidade Estadual do Norte Fluminense Darcy Ribeiro (UENF), registered under protocol 419-2017.

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Figures

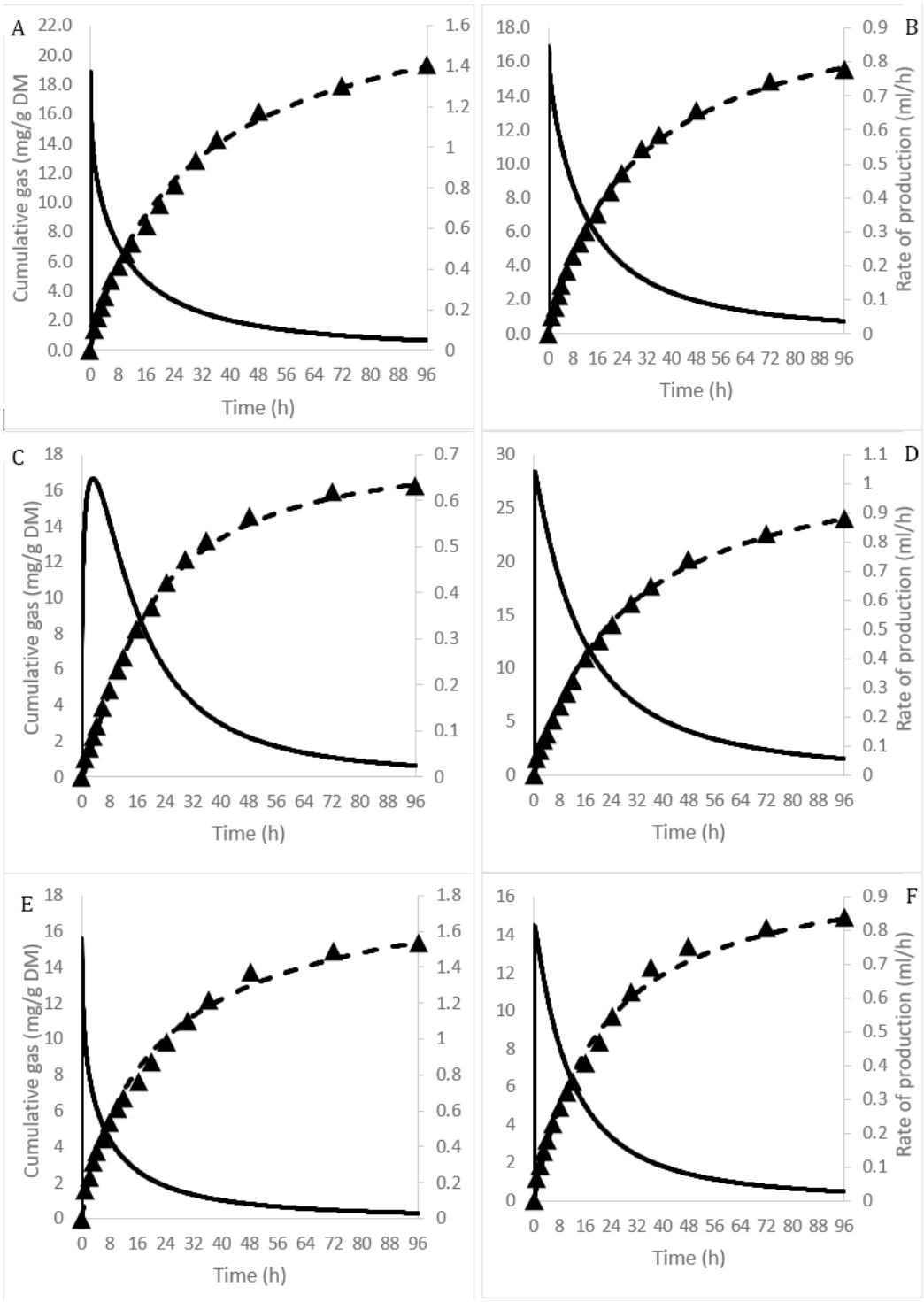

Figure 1

Cumulative gas production and rate of gas production profiles from pearl millet, corn, and sorghum silage with or not additives. On panel (1A) pearl millet without additive; (1B) pearl millet with additive; (1C) corn

without additive; (1D) corn with additive; (1E) sorghum without additive; and (1F) sorghum with additive [Lactobacillus plantarum (2.5 × 1010 cfu/g) and Propionibacterium acidipropionici (2.5 × 1010 cfu/g)].

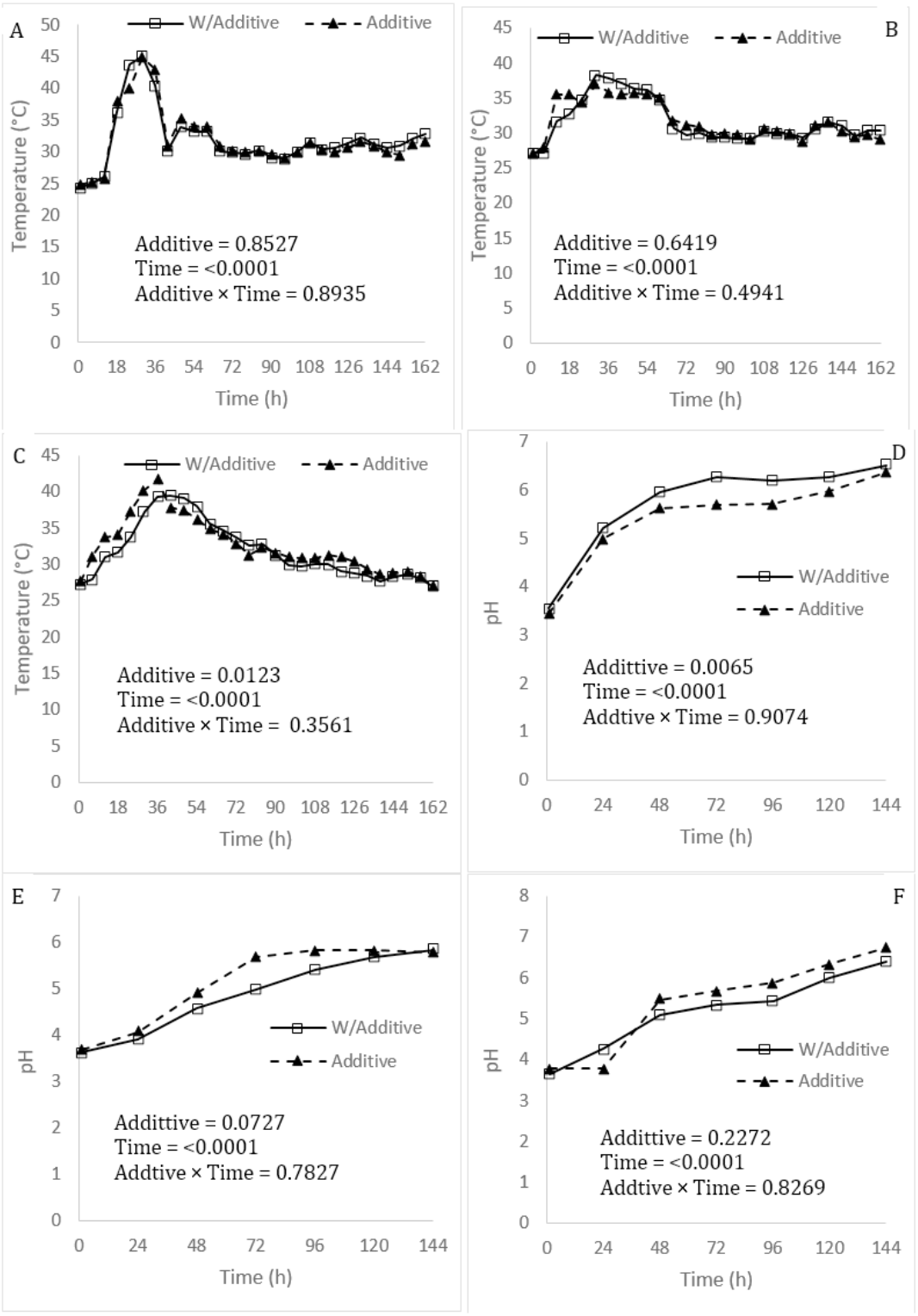

Figure 2

Aerobic stability of pearl millet, corn, and sorghum silage with or not additive. On panel (1A) temperature of pearl millet silage; (1B) temperature of corn silage; (1C) temperature of sorghum silage; (1D) pH of pearl millet silage; (1E) pH of corn silage; and (1F) pH of sorghum silage.