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AEROBIC STABILITY OF TIFTON 85 SILAGE WITH AND WITHOUT PRE-DRYING IN THE SUN

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Abstract

The objective of this study was to evaluate pH, ammoniacal nitrogen, and aerobic stability of silage of Tifton 85 grass silage with two dry matter contents at different silos opening times. The experimental design was completely randomized, in a subdivided plots scheme, in which the silages constituted the plots and aerobic exposure times the subplots, with four replications. To verify the aerobic stability of the silages, the temperature and pH were analyzed at seven hours after the silos were opened (1, 24, 48, 72, 96, 120, and 144 hours). The pH reached adequate levels for conservation only after 90 days of fermentation for the silages with and without pre-drying in the sun. Ammoniacal nitrogen remained below the recommended limits in both silages. As for the silage temperature, no loss of aerobic stability was observed. However, the observed pH revealed a break instability after 72 hours when the silos were opened at 28 days, with no changes for the remaining silage periods. It is possible to obtain suitable silages from Tifton 85 with or without pre-warming in the sun, however, a minimum fermentation period of 90 days should be adopted. The studied silages presented high aerobic stability, but when kept silage for only 28 days, they should be consumed by the animals within 48 hours after the supply.

Keywords: Ammonia Nitrogen. Cynodon. pH. Temperature.

1. Introduction

The seasonality of forage production is one of the obstacles to the development of Brazilian livestock (Santos et al. 2014). As an alternative to minimize the damages caused by the oscillations in forage production, we have the techniques of forage conservation, among which we highlight the anaerobic conservation through silage (Gayer et al. 2019).

This technique of conservation is one of the most widespread for the conservation of fodder, and even for tropical grasses, with or without the use of additives, has shown to be promising (Neres et al. 2013). The conservation of fodder in the form of silage has the principle of anaerobic fermentation, producing organic acids, promoting a drop in pH to values lower than 4.2, consequently preserving the quality of silage material (McDonald et al. 1991). A pH range indicated for well-preserved silages is between 3.6 and 4.2 (Moura et al. 2016).

A great diversity of forages can be conserved in the form of silage, provided they have a content of dry matter of 30% and 35%, soluble carbohydrate contents of 8 to 12% of MS, and low buffering power

(McDonald et al. 1991). These include Tifton 85 grass, which is a forage of high nutritional value, recommended for grazing, fencing, and silage (Santos et al. 2010).

However, commonly the forage mentioned above does not present the recommended dry matter contents for silage, requiring the use of wilting. Wilting or pre-drying makes it possible to ensilage the forage harvested with low dry matter content in a process in which the undesirable fermentations are controlled by raising the osmotic pressure obtained through the initial dehydration of the forage (Bernardes et al. 2018). The adoption of this technique prevents undesirable fermentations by raising the osmotic pressure inside the silo and also prevents the deterioration of the silage during fermentation or after the opening of the silos (Neres et al. 2013).

After opening the silos, the anaerobic environment inside the silo is broken and a series of transformations in the silage mass is initiated. Among these transformations, the action of yeasts that oxidize the organic acids preservatives of the silage is highlighted, triggering the aerobic degradation and, later, provoking the elevation of the pH and increase of the temperature that soon favors the growth of fungi. Thus, the evaluation of the aerobic stability of silages is performed by daily monitoring of the oscillations in the temperature and pH of the material exposed to air.

There are still few studies in the literature regarding the aerobic stability of wilted Tifton 85 silages. In this context, the present study aimed to determine the fermentation profile and to study the aerobic stability of Tifton 85 silage, silage with and without pre-drying to the sun during three fermentation periods.

2. Material and Methods

The experiment was conducted at a dairy farm located in the municipality of Santa Helena - PR and at the Laboratory of Animal Nutrition, State University of Western Paraná - *Campus* Marechal Cândido Rondon. For the study of the fermentation profile, a completely randomized design was used in subdivided plots (0, 28, 56, and 112 days after silage) allocated to the subplots and with the presence and absence of pre-drying in the sun allocated to the plots. For the test, the use of four replicates was used, totaling 32 experimental units, of which eight corresponded to the in natura materials, while the remaining 24 experimental units were made the experimental silos.

The Tifton 85 grass was harvested in a field destined to produce hay and silage with 32 days of regrowth and 30 cm high. A trapezoidal forage harvester, model Taarup was used to cut the plants, which was done at a height of 5 cm from the soil surface and the harvested forage was chopped into particles of approximately 5 cm and homogenized. For dehydration, part of the chopped forage was laid in the sun for two hours on a clean canvas forming a 10 cm high layer which was stirred twice every 45 minutes for 2 hours.

At harvest time, Tifton 85 grass had 22.5% dry matter (DM), while at the time of silage it had 26.0% DM and pre-dried Tifton 85 grass had 32.1% of DM. For the determination of the DM contents for each material, six random samplings were carried out with subsequent oven drying at 55°C with forced air circulation for 72 hours.

The experimental silos were made with PVC pipes 50 cm high and 10 cm in diameter. In each silo, 1.7 kg of shredded Tifton 85 grass were conditioned for the treatments without pre-drying, and 1.5 kg of Tifton 85 ground grass for the pre-drying treatments, respectively. The material was compacted, and the silos capped with caps equipped with Bunsen-type valves for the free escape of gases. The obtained densities were 432 and 380 kg.m^{-3,} which are lower than those recommended for the silages (McDonald et al. 1991), but they are due to the difficulty of compaction found during the assembly of the test.

At 0, 28, 56, and 112 days after silage the silos were opened for sampling, discarding the top and bottom of the silage. At each sampling, the values of pH and temperature and ammoniacal nitrogen (NH₃-N) of the silage were determined. The pH was determined using a potentiometer according to the methodology described by Cherney and Cherney (2003) and the silage temperature with a digital spit thermometer. Subsequently NH₃-N was determined by the methodology of Bolsen et al. (1992).

In the determination of the aerobic stability, after the opening of the silos, for each experimental unit a sample of 500 g was conditioned in plastic trays. For 144 hours, at 24 hours intervals, were measured the

silage temperature, the environment, and the pH of the silage exposed to air. The temperature and pH oscillations were measured to detect the interval between the opening of the silos and the breakdown of the aerobic stability of the silages.

In all pH determinations, a potentiometer was used in the aqueous extract formed by a fraction of 25 g of sample mixed to 450 mL of deionized water according to the methodology described by Cherney and Cherney (2003). Temperatures throughout the experimental period were measured with a digital spit thermometer.

It was considered as a breach of aerobic stability to elevate the temperature by 1°C above room temperature (Amaral et al. 2007) and to raise the pH by 0.2 units above the values observed at the time of opening of the silos (Jobim et al. 2007). The data obtained for pH and NH₃-N were submitted to analysis of variance and when it was found significant the absence and presence of pre-drying in the sun were compared by Fischer's F test, while the sampling times were studied using regression analysis, testing the linear (1st Degree) and quadratic (2nd Degree) models and considering the absence of significance for the regression deviations. The largest coefficient of determination (R²) was used to choose the model. The significance of the coefficients of the 1st (b1) and 2nd (b1 and b2) equations of the regression models selected for each studied variable were tested by Student's t-test.

The data obtained during the aerobic stability study were submitted to analysis of variance to obtain the coefficients of variation of the plots (CV1), subplots (CV2), and sub-subplots (CV3). As the study aimed to study the aerobic stability of silage based on reference values reported in the literature, no statistical tests were applied. Thus, for the variables studied in the evaluation of aerobic stability, the discussion was based on numerical values about established standards (pH and temperature) and not in the function of statistical differences.

3. Results and Discussion

For pH values, there was a significant effect of the interaction of the factors, and pH values for Tifton 85 silages with and without pre-drying in the sun adjusted to the quadratic regression model (Figure 1). The pH values decreased to 91 and 90 days after ensiling for Tifton 85 silages with and without pre-drying in the sun, respectively, with subsequent increase. At the time of ensiling the pre-dried Tifton 85 was found to have a higher pH (5.93) than the non-pre-dried Tifton 85 (5.40). During the fermentation period, the amplitude was reduced but remained up to 112 days (Figure 1).

The results obtained are consistent with those reported by Jobim et al. (2007), who point out that wilted silage, usually presents pH values above 4.2. Also, when referring to grass silages, it can be said that the pH reduction is hindered due to the low concentration of soluble carbohydrates since the low pH after ensiling is indicative of desirable fermentation (Santos et al. 2014).

The drop in pH is due to the action of lactic acid bacteria (Muck et al. 2018), which use soluble carbohydrates (such as those present in corn bran) to produce lactic acid (Kung Jr et al. 2018). Simultaneously with the growth of lactic acid bacteria, the development of undesirable microorganisms such as those of the genus *Clostridium* is inhibited (Driehuis et al. 2018).

The lower concentration of soluble carbohydrates and the increase in osmotic pressure reduce the fermentation intensity, resulting in a lower production of acids. This behavior indicates that these silages can reach fermentation stability with higher pH values because elevations in pH values with increasing dry matter contents in silages of grasses of the genus *Cynodon* were also observed by Quaresma et al. (2010).



Figure 1. pH values of Tifton 85 silage with (♦) and without (◊) pre-drying in the sun at the time of silage and with three opening times of the silos.

One of the desirable characteristics in the production of good quality silages is the rapid reduction of pH, which was not observed in this experiment. After ensiling, reductions of 0.62 (without pre-drying) and 0.94 (with pre-drying) were observed from the silage moment for the 28 days of fermentation. However, the pH values observed in the silage with 28 days of fermentation are not sufficient to ensure the conservation, because according to Moura et al. (2016), well-preserved silages present pH ranging from 3.6 to 4.2. The lowest pH values (4.2) were obtained only after 91 and 90 days of fermentation for the Tifton 85 silages with and without pre-wilting, respectively. Quaresma et al. (2010) obtained pH values of 4.87 for wilted Tifton 85 silage. The reduction of pH is not easily obtained, since grasses generally have low concentrations of soluble carbohydrates and high buffer capacity, preventing the rapid decrease of pH.

Regarding the NH₃-N of the silages, there was a significant effect of the interaction of the factors studied. At the time of ensiling, NH₃-N contents were similar among silage materials, however, from the 28 days of fermentation the silage made with Tifton 85 ensiled without prewarming showed higher NH₃-N contents than Tifton 85 silage pre-wilting. During the fermentation period a linear increase in the NH₃-N content of Tifton 85 silage was observed without pre-wilting, so that each day of the fermentation period an increase of 0.77% in the content of NH₃-N as a percentage of the total N of the silage, while in the silage obtained with the wilted Tifton 85, the NH₃-N contents presented a linear reduction of 0.53% at each day of the ensilage period (Figure 2).

NH₃-N is the result of plant and microbial proteolysis that occurs inside the silo (Kung Jr et al. 2018) or after aerobic exposure. This biochemical activity begins shortly after cutting the forage and lasts until the predominance of lactic fermentation (Woolford 1984), and the reduction of the pH of the silages below 4.2 so that acidic conditions are established inside the silo. Silages with levels of NH₃-N greater than 10% to 15% of the total N indicate intense proteolysis inside the silo, resulting mainly from the action of *Clostridium* (Kung Jr et al. 2018). Its evaluation is fundamental in silages because together with other non-nitrogen compounds, NH₃-N interferes in ruminal metabolism and the regulation of consumption in ruminants (Grant and Ferrareto 2018). Although increases in NH3-N were observed in all silages, their quality was preserved according to Kung Jr et al. (2018).

The dry matter content of the ensiled materials was similar to that of the Tifton 85 ensiled without wilting, with dry matter content of 26.0%, while wilted Tifton 85 presented dry matter contents of 32.1%. The wilting is a feasible strategy to decrease moisture content in short grasses used for silage (Bernardes et al. 2018), increasing the quality of obtained silages. Also, achieving a rapid wilting in the field is essential for reducing dry matter and nutritive value losses (Boreani et al. 2018) and developing deteriorators microorganisms (Driehuis et al. 2018). Increased DM decreases the *Clostridium* populations evidencing that the higher DM content helps in the control of these agents (Gayer et al. 2019). This fact is related to the predrying of the materials, which through the wilting of the ryegrass to the field caused a reduction of humidity and consequently a reduction of the presence of these microorganisms (Gentu et al. 2018).



Figure 2. NH₃-N values (% N Total) in Tifton 85 silage with (♦) and without (◊) pre-drying in the sun at the time of silage and with three opening times of the silos.

The wilting of forages limits the proteolysis of the material in the silo (McDonald et al. 2010), which is caused by the presence of microorganisms *Clostridium* since they are sensitive to high osmotic pressures, caused by the DM and absorption values, and for the relatively acidic conditions (pH <4.2) (Tavares et al. 2009). The silages obtained showed levels of NH₃-N consistent with those obtained by Quaresma et al. (2010) who obtained NH₃-N values of 7.34% for Tifton 85 silages.

Considering the results obtained for NH₃-N, the silages obtained can be classified as very good, as they have NH₃-N values below 10%, as reported by Pacheco et al. (2014). The results obtained demonstrate that the fermentations that occurred in the experimental silos of this study were adequate. According to Valeriano et al. (2009), the rapid drop in the pH of silages prevents the growth of microorganisms that degrade proteins, such as Enterobacteria and Clostridium, justifying the absence of an increase in the NH₃-N content of the studied silages.

The silages studied did not present a breakdown of the aerobic stability about the temperature, because the highest elevations of the silage temperature about the room temperature were observed at the moment of opening of the silos (Table 1). During the period of monitoring of aerobic stability, all temperatures measured in the silages were very close to or below the room temperature. In this way, all the silages studied presented aerobic stability for the temperature, because at no time after the opening of the silos the silages presented elevation of more than 1°C about the room temperature, considered as the limit for the breakdown of aerobic stability (Amaral et al. 2007).

czposure up	10 144 110013.							
Hours of – exposure	28 days		56 days		112 days			
	Without wilting	With wilting	Without wilting	With wilting	Without wilting	With wilting		
0	0.35	0.23	-1.43	-1.87	-0.58	-1.18		
24	-0.03	0.13	-0.07	-0.13	-0.87	-0.98		
48	-0.42	0.03	-0.68	-0.52	-0.97	-0.87		
72	-1.48	-0.50	-1.68	-1.57	-0.57	-0.40		
96	-2.33	-0.83	-0.47	-0.47	-0.50	-0.58		
120	-1.43	-0.38	-0.87	-0.68	-1.78	-1.67		
144	-1.03	-0.42	-0.82	-0.45	-0.78	-0.32		
CV1(%)	22.35							
CV2(%)	20.59							
CV3(%)	36.76							

Table 1. Oscillations relative to the room temperature in the temperatures of Tifton 85 silages with or without wilting in the sun, in three opening times of the silos (28, 56, and 112 days) after the aerobic exposure up to 144 hours.

CV1: coefficient of variation of plots (with and without wilting of Tifton 85); CV2: coefficient of variation of subplots (opening times of silos) and CV3: coefficient of variation of sub-subplots (hours of aerobic exposure).

The evaluation of the breakdown of aerobic stability in silages is relevant because the anaerobic environment of a silo is responsible for its conservation after the opening aerobic (Bernardes et al. 2009). After the opening of the silos, the breakdown of the aerobic stability due to the air penetration in the silo causes a decrease of the nutritive value with the aerobic deterioration of the ensiled material (Rezende et al. 2011). This process is due to the multiplication of the aerobic deteriorating microorganisms that were latent during the anaerobic phase of silage (Bernardes et al. 2009). In this study, about the silage temperature, it can be inferred that the dehydration period adopted was adequate, because it allowed to obtain silages that were stable after exposure to air (Table 1).

In the case of pH values, silage submitted to only 28 days of fermentation showed a breakdown of aerobic stability after 72 hours of exposure to air (Table 2), reaching pH values higher than 0.25 and 0.22 higher than observed when the silos are opened. The other silages, with silage periods of 56 and 112 days, did not exceed 0.2 pH values above the pH value at the opening of the silos, being silages with high aerobic stability. The pH values observed at the time of opening of silos also reflect that the silages obtained could reach values of aerobic stability with higher pH values (Quaresma et al. 2010).

Hours of — exposure	28 days		56 days		112 days			
	Without wilting	With wilting	Without wilting	With wilting	Without wilting	With wilting		
0	0.00	0.00	0.07	0.05	-0.09	-0.11		
24	0.08	0.05	0.19	0.03	-0.05	-0.15		
48	0.17	0.10	0.05	0.14	-0.04	-0.03		
72	0.25	0.22	-0.18	-0.11	0.11	-0.03		
96	0.34	0.35	-0.13	-0.08	0.01	0.05		
120	0.38	0.30	-0.15	-0.06	0.04	0.01		
144	0.42	0.25	-0.11	-0.05	0.07	0.08		
CV1(%)	25.11							
CV2(%)	65.66							
CV3(%)	22.39							

Table 2. Oscillations relative to the opening time of the silos at pH values of Tifton 85 silages with or without sun wilting in three opening times of the silos (28, 56, and 112 days) after aerobic exposure up to 144 hours.

CV1: coefficient of variation of plots (with and without wilting of Tifton 85); CV2: coefficient of variation of subplots (opening times of silos) and CV3: coefficient of variation of sub-subplots (hours of aerobic exposure).

The breakdown of aerobic stability revealed between 48 and 72 hours of air exposure to the silages with 28 days of fermentation confirms that the same when offered to the animals should be consumed preferentially in 48 hours. This recommendation is made based on the knowledge that the aerobic stability breakdown reveals the aerobic deterioration of these silages and also the risk of proliferation of potentially pathogenic or undesirable microorganisms (Driehuis et al. 2008). Therefore, the consumption of these silages after the breakage of their aerobic stability by the animals would negatively affect their performance.

The measurement of pH values during studies on aerobic stability of silages is relevant because the increase in pH values during the evaluation in aerobiosis is due to the consumption of the organic acids that preserve the silage (Ávila et al. 2012). The main acid consumed especially by yeasts is lactic acid, produced during anaerobic fermentation. The consumption of this acid causes an increase in pH and favors the development of deteriorating microorganisms (Gayer et al. 2019). The presence of these reduces the nutritional value of silage and inhibits the consumption of dry matter by animals (Ávila et al. 2012). Also, microbial growth, in turn, can trigger the contamination of silage by toxic products that can be produced by fungi and other bacteria, reducing the sanitary quality of the preserved food, and causing damages to the health of the animals (Driehuis et al. 2008).

4. Conclusions

The pre-drying in the sun increased the dry matter content of the Tifton 85 allowing to obtain a silage with values of pH and NH₃-N suitable for the conservation of material and feed of the animals. The silages made with Tifton 85, submitted or not to the wilting in the sun for two hours and silage by 28, 56 and 112 days of fermentation presented high aerobic stability, however, when kept silage for only 28 days, the silages should be consumed by the animals within 48 hours of delivery.

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