



## Additives in ensiling palisade grass managed under grazing intensities

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**ABSTRACT.** Conservation of summer forage excess represents a management strategy to meet animals' needs for dry matter in the shortage period, but has been poorly studied. Silage can be used for this purpose. This study analyzed the production of palisade grass silage from pasture subjected to different grazing intensities with and without additive, determining losses by gases and effluents and chemical composition of silage. The experiment was a 4 x 3 factorial completely randomized design, with four replications. The factors were: 1<sup>st</sup> – herbage allowance of 5% (5 kg dry matter 100 kg<sup>-1</sup> of animal weight day<sup>-1</sup>), 10, 15 and 20%. The pasture was managed under rotational stocking with 35-day grazing cycles (7 days of occupation and 28 days of rest) and 2<sup>nd</sup> - additives: a) control; b) citrus pulp pellets; c) biological inoculant for grass silage. The forage of palisade grass harvested from pastures subjected to low-intensity grazing showed quantitative and qualitative characteristics for ensiling. However, high humidity and low fermentable carbohydrate require the use of additive, favor the fermentation process and increase the nutritional quality of silage.

**Keywords:** inoculant, herbage allowance, citrus pulp, silage, experimental silo.

### Aditivos na ensilagem de capim-marandu manejado em intensidades de pastejo

**RESUMO.** Técnicas de conservação do excedente de forragem do verão constituem estratégia de manejo do pasto para suprir as necessidades dos animais em massa seca no período de escassez, porém pouco pesquisadas. A silagem pode ser utilizada para esse fim. Estudou-se a produção de silagem de capim Marandu, proveniente de pastos submetidos às intensidades de pastejo, com e sem aditivo, determinando-se as perdas por gases e efluentes e a composição química e bromatológica da silagem. O delineamento experimental foi inteiramente ao acaso e o experimento em fatorial 4x3, com quatro repetições. Os fatores foram: 1<sup>o</sup> – ofertas de forragem de 5% (5 kg de massa seca 100 kg<sup>-1</sup> de peso animal dia<sup>-1</sup>), 10, 15 e 20%. A pastagem foi manejada em lotação rotativa, com ciclos de pastejo de 35 dias (7 dias de ocupação e 28 dias de descanso) e 2<sup>o</sup> – aditivos: a) controle; b) polpa cítrica peletizada; c) inoculante biológico para silagens de capim. Verificou-se que a forragem de capim-marandu colhida de pastos submetidos à baixa intensidade de pastejo apresentou características quantitativas e qualitativas para ensilagem. Porém, a alta umidade e o baixo teor de carboidratos fermentescíveis tornam a utilização de aditivo necessária e favorável ao processo fermentativo e a qualidade nutricional da silagem.

**Palavras-chave:** inoculante, oferta de forragem, polpa cítrica, silagem, silo experimental.

### Introduction

Is a component of a complex ecosystem, which provides substrates to the animals, but presents qualitative and quantitative variations throughout the year, mainly influenced by biotic and abiotic factors and by management techniques (Moreira, Prado, Cecato, Wada & Mizubuti 2004). These elements promote seasonality in the production of forage crops, concentrating approximately 80% of production in the rainy period and 20% in the dry period and may vary with the intensification level used in the pasture management (Euclides, Flores, Medeiros & Oliveira 2007; Herling et al., 2011).

Aiming to achieve higher productive levels, researchers have been encouraged to develop solutions to meet the growing demand for forage, especially during the dry period. The seasonal production of forage crops requires the use of management strategies as deferred pasture and/or forage conservation practices during the growth period of these plants in order to meet the qualitative and quantitative needs along the shortage period, thus minimizing the effects of seasonal food availability (Lucato Junior & Paula, 2008; Santos et al., 2010).

In recent years, the importance of grass silage increased significantly, given the great advances in

research validating its nutritional quality. In the ensiling process, to obtain the desired fermentation, it is essential the anaerobic conditions inside the silo, achieved mainly by the effective compaction. The ease of compaction of the material depends on the dry matter content in the forage, the particle size at the time of ensiling (Santos et al., 2010), which facilitates the ensiling process, since it allows higher density of transport of the collected material to the storage location, but also increases the efficiency of the compaction process and allows a better anaerobic fermentation (Neumann et al., 2007).

Ensiling grasses may require the use of additives because of their interference in reducing losses and in getting a better nutritional value of the silage, considering the excess moisture or low fermentable carbohydrate content in harvested forage. Kleinschmit and Kung (2006) reported that the use of *Lactobacillus* promoted a reduction in pH, lactic acid concentration and number of yeasts, as well as increased the concentration of acetic acid and a greater stability of the inoculated silages; however, many of the results on the use of microbial additives are controversial (Muck, 2010).

Among the sources of dry matter and fermentable carbohydrates, citrus pulp is a favorable additive in grass ensiling. Bernardes et al. (2005) observed its benefits in the fermentative and microbiological profile in palisade grass silage, and Rodrigues et al. (2005) found that its high soluble carbohydrate content provided higher content of these in ensiled mass, and improved the quality of fermentation and the nutritional value of the silage.

Considering the above, the goal of this study was to evaluate the production of silage of palisade grass from pastures subjected to different grazing intensities with the use of additives by means of determining losses by gases and effluents during anaerobic fermentation in the chemical composition of these silages, resulting from the morphological composition of the harvested forage.

## Material and methods

The experiment was conducted at the Animal Science Department, Faculty of Animal Science and Food Engineering, University of São Paulo - USP/FZEA in the municipality of Pirassununga, São Paulo State, Brazil (21° 59' S and 47°26' W, 634 m altitude). The climate is classified as subtropical Cwa (Köppen & Geiger, 1928), with rainfall and average annual temperature of 1,300 mm and 23°C. The soil is classified as dystrophic Dark Red Latosol (EMBRAPA, 2006).

The harvest of palisade grass [*Urochloa brizantha* (Stapf) Webster cv. Marandu] took place before the animals enter the pasture area managed by rotational stocking during the rainy period, with 35-day grazing periods, seven days of occupation and 28 days of rest. The adjustment of the stocking rate was held each grazing cycle, considering the treatments of herbage allowance.

The experiment was a 4 x 3 factorial completely randomized design, with four replications. The factors were: 1<sup>st</sup> - Herbage allowance (HA): 5% (5 kg dry matter 100 kg<sup>-1</sup> of animal weight day<sup>-1</sup>), 10, 15 and 20%; 2<sup>nd</sup> - additives: a) control; b) citrus pulp pellets at 7.5%; c) biological inoculant for grass silage. The adjustment of the paddock stocking was performed at each grazing cycle, considering the tester and regulators animals.

Herbage mass was harvested manually 5 cm from the ground in pre-grazing and then minced into medium particles of 1 to 2 cm and subjected to the treatments: citrus pulp pellets at 7.5% (75 kg ton<sup>-1</sup> forage) and biological inoculant (*Enterococcus faecium*, *Lactobacillus plantarum* and *Lactobacillus salivarius*, cellulase and hemicellulose at 5% - 250 g 50 L<sup>-1</sup> water) by spraying 1 L ton<sup>-1</sup> forage.

The material was ensiled in experimental silos made of plastic (PVC) with 10 cm diameter and 30 cm height, sealed by a cap (flow switch) at each end with Bunsen valves for outgassing at the top and rubber hoses for the flow of the effluent at the bottom.

Forage was compacted with wooden rafter using as criterion the accommodation of layers of approximately 5 cm thick, so that the pressure in each experimental silo was similar, allowing to obtain constant density of ensiled mass. We evaluated forage harvested in two grazing cycles, in February and March separately, with four repetitions, totaling 96 small silos for each grazing cycle.

The chemical composition of the forage at pre-ensiling, during the first and second grazing cycles, expressed the potential to be conserved as silage. At lower grazing intensities, the palisade grass presented the best characteristics for ensiling (Table 1).

The effluent produced during the fermentation was weighed every two days and the loss of gas during fermentation was determined indirectly after opening small silos at 90 days, by subtracting the final silage mass from the initial silage mass in the experimental silo added to effluent losses over the 90 days.

**Table 1.** Dry matter content (DM%) and chemical composition on a dry matter basis of neutral detergent fiber (NDF%), acid detergent fiber (ADF%), crude protein (CP%), pH, buffering capacity (BC%), soluble carbohydrates (SC%), calcium (Ca%) and phosphorus (P%) of palisade grass harvested in the first and second summer grazing cycle.

Treatments	HA*	DM	NDF	ADF	CP	pH	BC	SC
First grazing cycle								
Control		19.00	87.60	47.36	5.63	7.29	3.87	6.70
Inoculant	5	20.00	83.82	48.51	5.36	5.62	6.67	8.79
Citric Pulp		25.22	83.66	42.71	6.40	4.98	4.28	10.01
Control		19.22	87.71	46.83	5.86	7.06	3.80	7.59
Inoculant	10	23.58	84.05	47.70	4.58	6.60	5.50	6.90
Citric Pulp		21.24	80.75	41.83	6.31	5.09	4.32	13.74
Control		19.42	87.07	49.15	5.25	6.77	3.56	6.17
Inoculant	15	30.63	84.30	49.90	4.00	5.08	5.38	6.41
Citric Pulp		23.82	80.16	43.54	4.60	5.16	4.64	10.68
Control		18.86	88.51	49.59	4.14	7.39	2.88	5.16
Inoculant	20	30.27	81.55	51.25	4.85	6.45	5.45	7.42
Citric Pulp		29.94	82.36	44.14	5.27	6.13	5.04	9.50
Second grazing cycle								
Control		28.26	78.28	43.33	6.17	6.04	5.92	10.46
Inoculant	5	29.33	79.03	44.87	7.00	5.99	8.42	12.50
Citric Pulp		40.60	79.54	40.02	5.95	6.60	10.12	12.14
Control		29.39	81.17	46.15	5.08	6.01	6.24	9.61
Inoculant	10	33.95	80.76	48.05	5.29	5.78	9.04	10.97
Citric Pulp		41.87	77.53	39.58	5.77	5.35	9.08	14.07
Control		33.19	81.05	47.40	4.32	5.96	7.02	10.94
Inoculant	15	35.13	81.39	47.87	4.95	5.84	8.23	10.12
Citric Pulp		39.64	74.54	43.87	5.07	5.38	9.31	12.89
Control		29.28	77.81	44.85	6.37	6.20	7.68	10.03
Inoculant	20	32.71	80.82	51.14	4.36	5.99	7.56	10.98
Citric Pulp		35.71	75.31	45.38	4.75	6.13	12.35	14.51

\*HA: Herbage allowance.

After opening the small silos, we observed the presence of spoiled silage (contaminated with fungi, molds, coloration and smell), which was separated, while the silage considered as good was homogenized and frozen and then sampled, dried in a forced air circulation (55°C) and ground to determine the chemical composition.

Analysis of neutral and acid detergent fiber were performed according to Goering and Van Soest (1970), and the crude protein analysis was carried out according to Kjeldahl method (AOAC, 2005). The total soluble carbohydrates (CHO) were determined according to Dubois, Gilles, Hamilton, Rebers and Smith (1956). The potential of hydrogen (pH), calcium (Ca) and phosphorus (P) were determined according to Silva and Queiroz (2002) and the buffering capacity (BC %) according to Playne and McDonald (1966). Short chain fatty acids were determined by gas chromatography (Sigma-Aldrich Co.), and lactic acid in the high performance liquid chromatography system for (HPLC) according to Wilson (1971).

Data were statistically analyzed using the PROC GLM procedure of SAS (2004). It was considered the

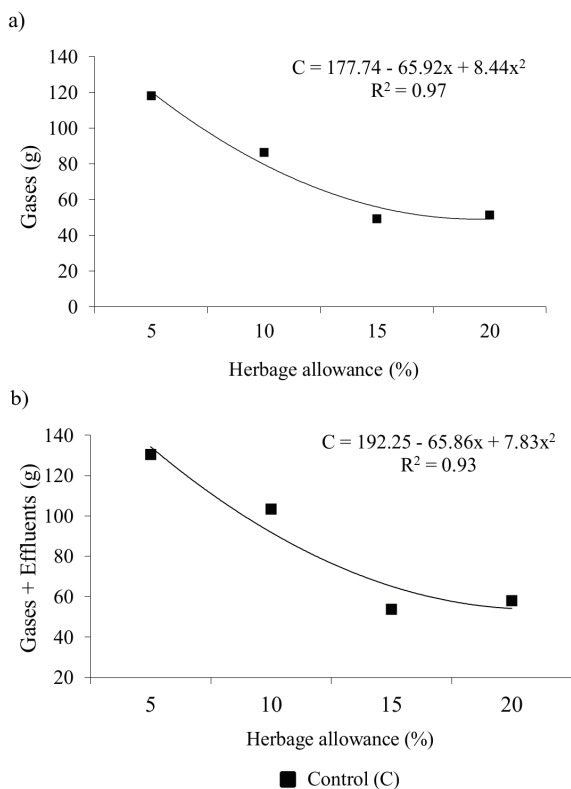
main effects of the 1<sup>st</sup> and 2<sup>nd</sup> factors as well as the interaction between them, for each period separately, and because of the significant interactions, we applied regression analysis to verify the behavior of each additive in terms of herbage allowance evaluated.

## Results and discussion

The pH readings of silage, for the first grazing cycle, with the additives inoculant ( $p < 0.01$ ) and citrus pulp ( $p < 0.01$ ) showed a downward linear behavior. In the second grazing cycle, the pH readings, considering the treatments control ( $p < 0.01$ ), inoculant ( $p < 0.01$ ) and citrus pulp ( $p < 0.01$ ) presented a quadratic behavior. For the treatments control, inoculant and citrus pulp, the lower pH readings would be obtained if the herbage mass was harvested at herbage allowance of 11.7; 10.0 and 12.5%, respectively.

The losses by gases and gases + effluents during fermentation were different ( $p < 0.05$ ) between herbage allowance only for the control in February. The behavior was quadratic with reduced losses according to herbage allowance (Figure 1). The results are sustained only by the decrease in buffering capacity (3.87 to 2.88), although the contents of dry matter and readily fermentable carbohydrates have been low and much lower than the other treatments (Table 1). The bacteria comprising the inoculant ferment sugars into lactic acid, resulting in less loss of nutrients and better silage quality (McDonald, 1981).

Bernardino, Garcia, Rocha, Souza and Pereira (2005) evaluated the effects of adding 10, 20, 30 and 40% coffee hulls to elephant grass (*Pennisetum purpureum* Schum.) and concluded that the addition of 20% coffee hulls ensured good conservation of silage without effluent production. Coffee hulls enrich the silage with dry matter, and also have effect retention of free water, eliminating some of the moisture. Rodrigues et al. (2005) stated that lower production of effluents by using citrus pulp pellets provides increased soluble carbohydrate concentration available in ensiled material, improves the fermentation and consequently reduces losses by gases. Tavares et al. (2009) observed that, with increasing dry matter by the addition of citrus pulp, effluent production decreased and ceased. These results are in agreement with those obtained in the present study, with no significant production of effluent.



**Figure 1.** Loss of gases (g) (G - g) and gases + effluent (GE - g), a and b respectively, for the control treatment in the first evaluation cycle.

No loss by gases was found for the forage harvested in the second grazing cycle and ensiled, and gas losses did not differ between treatments (Table 2). This behavior is explained by the higher dry matter content of the forage in the ensiling process (Table 1), compared to that harvested in February. The high moisture content in forage negatively interferes with the fermentation, promoting losses through unwanted fermentation and/or nutrient leaching by effluent percolation.

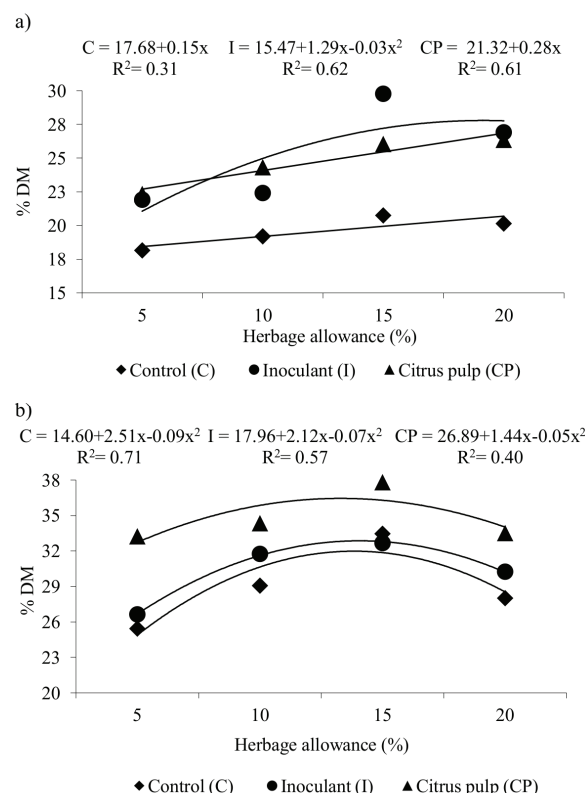
**Table 2.** Loss of gases (g) during the fermentation of forage harvested in the second grazing cycle.

Treatments	Herbage allowance (%)			
	5	10	15	20
	Gases (g)			
Control	26.7	28.0	19.9	25.0
Citrus Pulp	24.2	27.1	17.2	23.9
Inoculant	22.6	18.8	27.0	22.3
EPM	2.45	2.45	2.45	2.45

Mean values in the same column are not significantly different from each other ( $p > 0,05$ ).

The dry matter content of the forage has pronounced effects on its content in the silage. Although it is expected a lower dry matter content when including the inoculant, compared to citrus pulp, this behavior occurred only with forage pre-ensiling in the second cycle. There was a linear

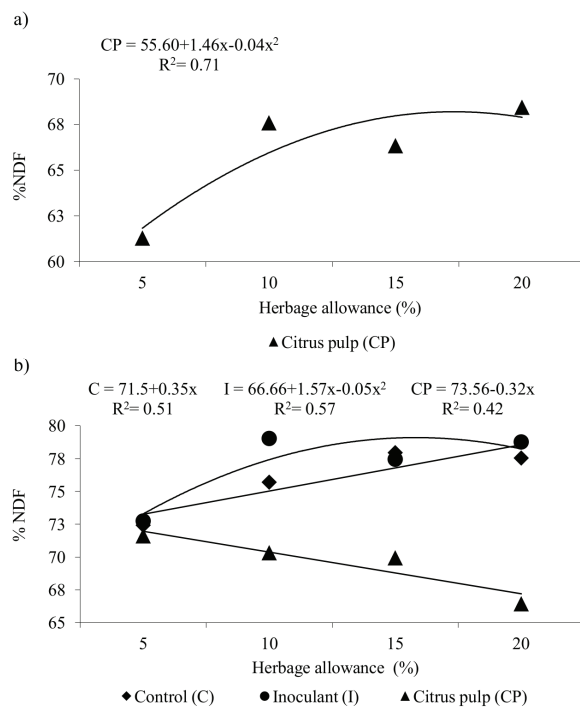
behavior for the treatments control ( $p < 0.01$ ) and citrus pulp ( $p < 0.01$ ), and quadratic for the inoculant ( $p < 0.05$ ) in the first grazing cycle, while in the second cycle, all treatments presented a quadratic behavior ( $p < 0.01$ ) (Figure 2). The dry matter content at pre-ensiling reflected in the contents of the respective silages. In the second cycle, as the ensiled forages have shown high counts of dry matter, there was no loss by effluent.



**Figure 2.** Dry matter (DM%) content of palisade grass silage, considering the treatments of herbage allowance and use of additives in the first (a) and second (b) grazing cycles.

The results obtained with the silages may be related to the morphological composition of the plant used for ensiling, with greater mass of forage and senescent material at the supply of 20% compared to the others. The neutral detergent fiber content in the first grazing cycle was affected by the treatments of supply, presenting a quadratic fit ( $p < 0.01$ ) with the use of citrus pulp. The average content of NDF in forage at pre-ensiling was 82.5%, ranging from 80.2 to 87.7% and, after the fermentation process, the values ranged from 62.5 to 67.5%. Whereas part of the cell wall can be broken down by enzymes and carbohydrates are fermented, the difference of 15 to 20 percentage units, between the pre- and post-ensiling, may have been used for this purpose. The forage harvested in the second cycle showed, on average, NDF content

varying between 76.7 (CP), 79.6 (c) and 80.5% (I). After 90 days of fermentation, the silage sampled showed a positive linear behavior ( $p < 0.01$ ) for the control and negative ( $p < 0.01$ ) for citrus pulp, and quadratic effect ( $p < 0.05$ ) for the inoculant, with maximum point at herbage allowance of 15.7% (Figure 3). The silage showed average NDF contents ranging from 67.5 to 77.5%. The range of variation in silage with forage of the second grazing cycle was higher, once the observed behavior may reflect the growing presence of green leaves, stem and green sheaths and senescent material in the composition of the ensiled material with increasing levels of herbage allowance.

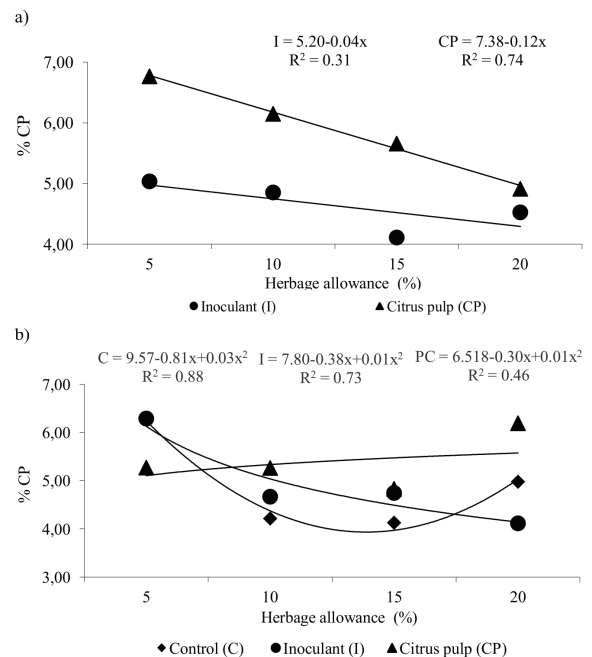


**Figure 3.** Neutral detergent fiber (NDF) in palisade grass silage, considering the treatments of herbage allowance and use of additives in the first (a) and second (b) grazing cycles.

Values of the acid detergent fiber content, considering the first and second grazing cycles, varied from 43.1 and 42.2 (CP); 48.2 and 45.4 (C), and 49.3 and 48.0% (I) in the pre-ensiling forage. The palisade grass silage harvested in the first cycle averaged 44.2% with citrus pulp, differing significantly from the control (49.1%) and from the silage with inoculant (48.2%), which were not significantly different from each other, while in the forage silage of the second cycle, all silages differed from each other ( $p < 0.05$ ) with 48.1, 45.8 and 43.3%, respectively, for the treatments control, inoculant and citrus pulp. According to Loures et al. (2005), when evaluated the chemical composition of Tanzania grass silage, the ADF content in the control was 44.4%, close to the

value obtained in this study. ADF has not been practically used carbohydrate source for fermentation, considering the values before and after ensiling. This did not occur with the NDF, rich source of hemicellulose that can be used as a source of fermentable carbohydrate in the process.

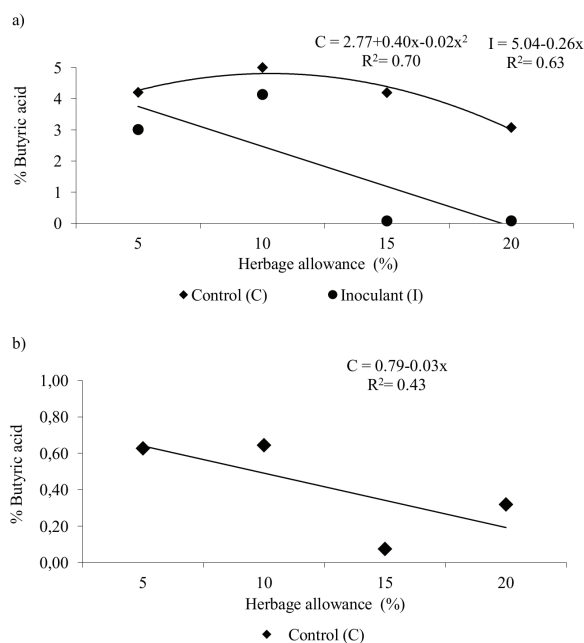
The crude protein content of the silage with inoculant ( $p < 0.05$ ) and citrus pulp ( $p < 0.01$ ) of the first grazing cycle showed negative linear fit. In the second cycle, all treatments presented quadratic fit, the minimum points at the supplies of 13.5; 19.0 and 15.0% for the treatments control, inoculant and citrus pulp, respectively (Figure 4). Higher values of protein content were observed in silage of pastures under higher grazing intensity, except for the inclusion of CP to the ensiled mass from pasture managed under less intensive grazing (HA = 20%). Rodrigues, Borgatti, Gomes, Passini and Meyer (2005) evaluated the effects of adding increasing levels of citrus pulp on fermentative quality and on nutritional value of elephant grass silage and registered an average CP content of 6.8%, which are consistent with the results obtained in this study. Similar crude protein content between pre- and post-ensiling assumes low use of protein sources in the fermentation process.



**Figure 4.** Crude protein (CP) content in palisade grass silage, considering the treatments of herbage allowance and use of additives in the first (a) and second (b) grazing cycles.

The butyric acid content of silage with inoculant ( $p < 0.01$ ) of the first grazing cycle showed a decreasing linear fit of the order of 0.26% for every 1% of the supply, while for the control ( $p < 0.05$ ), we observed a quadratic fit with the asymptote for the supply of 10%.

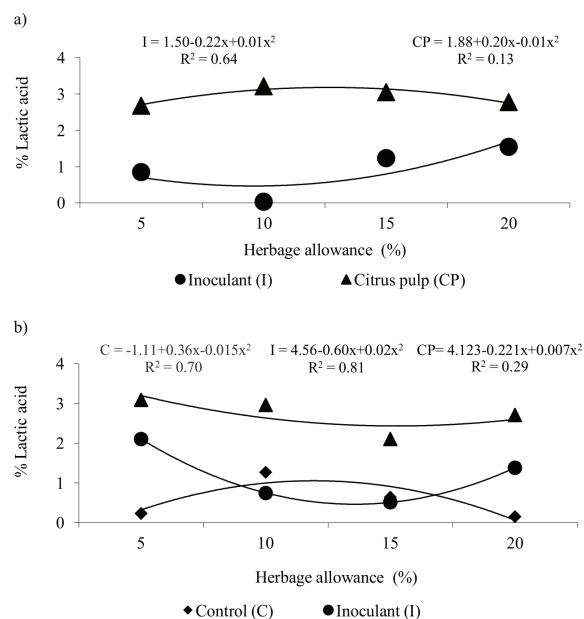
In the second cycle, the control ( $p < 0.01$ ) showed a decreasing linear fit of the order of 0.03% for every 1% of herbage allowance. Low content of forage dry matter of the first grazing cycle provided a favorable environment for butyric fermentation, especially that originating from pastures managed under higher grazing intensity (HA = 5 and 10%). Carvalho et al. (2008) observed higher butyrate content in elephant grass with lower content of dry matter, similarly to the present study. In the second forage grazing cycle, with higher dry matter content, the percentages of butyric acid were lower, but still high in the pastures managed with high stocking rates (HA = 5 and 10%), and this trend may have directly interfered with the percentage content of butyric acid, inhibiting the growth of microorganisms responsible for the production of this acid (Figure 5).



**Figure 5.** Butyric acid of palisade grass silage, considering the treatments of herbage allowance and use of additives in the first (a) and second (b) grazing cycles.

In turn, the lactic acid content in silage of forage harvested in the first cycle presented a quadratic fit, when applied the inoculant ( $p < 0.01$ ) and citrus pulp ( $p < 0.01$ ). In the second cycle, all treatments presented a quadratic behavior (Figure 6). Santos et al. (2008) verified that the inoculation increased lactic acid content and decreased the contents of acetic acid and butyric acid. In *Pennisetum hybridum* cv. Paraíso at 100 days of age, Ferrari Junior, Paulino, Possenti and Lucenas (2009) found that the percentages of lactic acid in the silage increased with the addition of citrus pulp up to 10%. The citrus pulp applied in ensiled forage in the two grazing cycles of this study was an important additive in ensiling palisade grass, as it maintained high

lactate contents in silage. In guinea grass (*Panicum maximum* Jacq), Penteado et al. (2007) found that inoculation with *L. plantarum* from epiphytic microbiota improves the fermentation profile, relative to the contents of lactic acid, pH,  $\text{NH}_3$  and acetic acid, and lower losses of dry matter in silage. There was a lower content of lactic acid in the treatments without additives. This acid is related to the presence of beneficial microorganisms in anaerobic environments with high content of dry matter and soluble carbohydrates.



**Figure 6.** Lactic acid in the palisade grass silage considering the treatments of herbage allowance and use of additives in the first (a) and second (b) grazing cycles.

## Conclusion

Although it has lower protein content, the forage harvested from pasture under more lenient management has better characteristics for fermentation.

The inclusion of citrus pulp and inoculant is feasible and necessary for fermentation of forage harvested from lower pastures, thus increasing the dry matter and fermentable carbohydrates content, important for lactic acid production.

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