**LUCIANO SARAIVA SANTOS**

# EFFECTS OF ADDITIVES ON FERMENTATION PROFILE AND *IN SITU* RUMEN DEGRADABILITY OF ELEPHANT GRASS SILAGE CV. BRS CAPIAÇU

Dissertation submitted to the Animal Science Graduate Program of the Universidade Federal de Viçosa in partial fulfillment of the requirements for the degree of *Magister Scientiae*.

Adviser: Polyana Pizzi Rotta

Co-advisers: Alex Lopes da Silva
Odilon Gomes Pereira

**VIÇOSA - MINAS GERAIS**
**2022**

**Ficha catalográfica elaborada pela Biblioteca Central da Universidade Federal de Viçosa - Campus Viçosa**

T
S237e
2022

Santos, Luciano Saraiva, 1996-
Effects of additives on fermentation profile and *in situ* rumen degradability of elephant grass silage cv. BRS Capiaçu / Luciano Saraiva Santos. – Viçosa, MG, 2022.
1 dissertação eletrônica (67 f.): il.

Texto em inglês.
Orientador: Polyana Pizzi Rotta.
Dissertação (mestrado) - Universidade Federal de Viçosa, Departamento de Zootecnia, 2022.
Inclui bibliografia.
DOI: https://doi.org/10.47328/ufvbbt.2023.073

1. Silagem. 2. Rações - Aditivos. 3. Rúmen - Fermentação. I. Rotta, Polyana Pizzi, 1987-. II. Universidade Federal de Viçosa. Departamento de Zootecnia. Programa de Pós-Graduação em Zootecnia. III. Título.

CDD 22. ed. 636.0862

Bibliotecário(a) responsável: Bruna Silva CRB-6/2552

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Dissertation submitted to the Animal Science Graduate Program of the Universidade Federal de Viçosa in partial fulfillment of the requirements for the degree of *Magister Scientiae*.

APPROVED: August 19, 2022.

Assent:

Luciano Saraiva Santos
Author

Polyana Pizzi Rotta
Adviser

To my grandfather, brother, and uncle (*In memory*) that I will always love.

## ACKNOWLEDGEMENTS

I want to thank my parents Maria Olindina and Irani Calixto for all the support, for believing in me in my ability and all the teachings, which always made me a better human being.

I need to thank my girlfriend Tatiana Brumano for always being by my side supporting me and always believing in me, even when I didn't believe in myself.

I want to thank my grandfather, uncle and brother (in memoriam) for the teachings, companionship, friendships and always encouraging me to study and try to be a better professional. All my dedication and commitment is dedicated to you.

I also want to thank my advisor Polyana Rotta for these two years of teaching and support. I learned a lot about research and had the opportunity to learn a lot about dairy cattle from her. No doubt she is a great professional.

I also want to thank Professor Alex for teaching me so much about statistics and for his advice and friendship throughout my Masters. I learned a lot from him about how to be an excellent professional.

I would like to thank my family for their support, encouragement and for always believing in me.

I thank Dr Domingos for always encouraging me, believing in my potential and teachings throughout my seven years at UFV.

Thanks to João, Poliana, Gabriel and Luis Henrique, my personal friends that I gained throughout my master's degree, for their friendship and support in carrying out my experiment.

I thank Bernardo Martins for his friendship, help during this study and support at all times.

This dissertation would not have been possible without the help of my interns and personal friends from the dairy laboratory: Dayanne Lima, Marina, Julia Lopes, Julia Mapa, Welington, Thaís and Vitória.

I would also like to thank my co-supervisor: Odilon Gomes for the analyzes carried out in the forage laboratory and thank Professor Adenilson Paiva for participating in the committee and contributing to the work.

Thanks to the Department of Animal Science, UEPE-GL and Família do Leite of Universidade Federal de Viçosa where I had the opportunity to develop a passion for research.

This study was financed in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – Brasil (CAPES) – Finance Code 001.

To the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq), for granting the scholarship.

*"It is on the way up that the cinnamon thickens"*
(Evandro Guedes)

## BIOGRAPHY

Luciano Saraiva Santos, son of Maria Olindina Saraiva dos Santos and Irani Calixto dos Santos, was born in Jabaquara, SP – Brazil on February 8, 1996. The grandson of a rural producer has always been interested in animal production.

He started Zootechnics in 2015 and in 2020 he became a zootechnician at the Federal University of Viçosa, Viçosa, MG – Brazil.

In August 2020, he started his master's degree in Animal Science with a specialization in ruminant nutrition at the Federal University of Viçosa, under the guidance of Dr. Polyana Pizzi Rotta. She submitted her dissertation to the committee on August 19, 2022.

## ABSTRACT

SANTOS, Luciano Saraiva, M.Sc., Universidade Federal de Viçosa, August, 2022. **Effects of additives on fermentation profile and *in situ* rumen degradability of elephant grass silage cv. BRS Capiaçu.** Adviser: Polyana Pizzi Rotta. Co-advisers: Alex Lopes da Silva and Odilon Gomes Pereira.

The objective of this work was to evaluate the effects of four doses of different bacterial additives or the inclusion of 8% ground corn grain in elephant grass cv. BRS Capiaçu on the silage fermentative profile, chemical composition, and *in situ* ruminal degradability. The experimental design was completely randomized, with treatments set up in a 6 × 3 factorial scheme (six additives × three fermentation periods) and three replications. The additives were 1) no inoculant (NI); 2) 0.5 g/ton of commercial product composed of *Lactobacillus buchneri* ($LBU_{0.5}$); 3) 1 g/ton of commercial product composed of *Lactobacillus buchneri* ($LBU_1$); 4) 1 g/ton of commercial product composed of *Lactobacillus plantarum* and *Pediococcus acidilactici* ($LPP_1$); 5) 2 g/ton of commercial product composed of *Lactobacillus plantarum* and *Pediococcus acidilactici* ($LPP_2$); and 6) inclusion of 8% ground corn grain (IGCG) in the silage, which was opened at 30, 60 and 90 d of fermentation. The silage opened at 60 d of fermentation was subjected to *in situ* rumen degradability procedures using three lactating Holstein cows fistulated in the rumen, allowing for the following incubation times (0, 3, 6, 12, 24, 30, 48, 72, 120 and 240 h). Silage with IGCG showed the highest levels of dry matter (DM), crude protein, and other extract and reductions in the neutral detergent fiber (NDF), acid detergent fiber and lignin. For the fermentation profile, silages containing IGCG showed the lowest pH, butyric acid, and ammoniacal nitrogen levels and the highest lactic acid levels. Water-soluble carbohydrates and total DM losses did not differ between treatments. Silages containing IGCG showed greater potential and effective degradability of DM and organic matter (OM), with a consequent increase in fraction "a" (soluble fraction) and a reduction in fraction "b" (potentially degradable insoluble fraction). The potential degradability of NDF showed the highest values, however, differing only from silages supplemented with $LBU_1$. The effective degradability of NDF and its fractions "C" (the degradation rate of the potentially degradable fraction) and "I" (the indigestible fraction) was not influenced by the use of additives. When the "D" fraction (the potentially degradable fraction of the NDF) was observed, the silage containing IGCG presented the highest value; however, it did not differ from the NI, $LPP_1$ and $LPP_2$ silages. DM and MO fraction "c" did not differ between additives. It is concluded that the silage containing IGCG presented the best fermentative

profile and the best nutritional value, providing increases in the ruminal degradability of DM and Capiaçu MO, improving the degradation of nutrients, and providing an alternative for the availability of nutrients in the rumen.

**Keywords:** Additives. BRS Capiaçu silage. Fermentation profile. *In situ* ruminal degradability. Nutritional value.

## RESUMO

SANTOS, Luciano Saraiva, M.Sc., Universidade Federal de Viçosa, agosto de 2022. **Efeitos de aditivos no perfil de fermentação e na degradabilidade ruminal *in situ* da silagem de capim-elefante cv. BRS Capiaçu.** Orientador: Polyana Pizzi Rotta. Coorientadores: Alex Lopes da Silva e Odilon Gomes Pereira.

O objetivo deste trabalho foi avaliar os efeitos de quatro doses de diferentes aditivos bacterianos ou inclusão de 8% de grão de milho moído em silagem de capim elefante cv. BRS Capiaçu, sobre o perfil fermentativo, composição química e degradabilidade ruminal *in situ*. O delineamento experimental utilizado foi inteiramente casualizado, com tratamentos em esquema fatorial 6 × 3 (seis aditivos × três períodos de fermentação) e três repetições. Os aditivos foram: 1) sem inoculante (NI); 2) 0,5 g/ton de produto comercial composto por *Lactobacillus buchneri* ($LBU_{0,5}$); 3) 1 g/ton de produto comercial composto por *Lactobacillus buchneri* ($LBU_1$); 4) 1 g/ton de produto comercial composto por *Lactobacillus plantarum* e *Pediococcus acidilactici* ($LPP_1$); 5) 2 g/ton de produto comercial composto por *Lactobacillus plantarum* e *Pediococcus acidilactici* ($LPP_2$); e 6) inclusão de 8% de grão de milho moído (IGCG) na silagem, aberta aos 30, 60 e 90 d de fermentação. A silagem aberta aos 60 dias de fermentação foi submetida aos procedimentos de degradabilidade ruminal *in situ*, utilizando três vacas holandesas lactantes fistuladas no rúmen, permitindo os seguintes tempos de incubação (0, 3, 6, 12, 24, 30, 48, 72, 120 e 240 h). Observou-se que a silagem com IGCG apresentou os maiores teores de matéria seca (MS), proteína bruta, extrato etéreo e reduções de fibra em detergente neutro (FDN), fibra em detergente ácido e lignina. Quanto ao perfil de fermentação, as silagens com IGCG apresentaram os menores teores de pH, ácido butírico e nitrogênio amoniacal, e os maiores teores de ácido lático. Os carboidratos solúveis em água e as perdas totais de MS não diferiram entre os tratamentos. As silagens com IGCG apresentaram maior degradabilidade potencial e efetiva da MS e da matéria orgânica (MO), com consequente aumento de sua fração "a" (fração solúvel) e redução da fração "b" (fração insolúvel potencialmente degradável). A degradabilidade potencial da FDN apresentou os maiores valores, entretanto diferindo apenas das silagens aditivadas com $LBU_1$. A degradabilidade efetiva da FDN e suas frações "C" (velocidade de degradação da fração potencialmente degradável) e "I" (fração indigerível) não foi influenciada pelo uso de aditivos. Aos observar a fração "D" (fração potencialmente degradável da FDN), a silagem com IGCG apresentou o maior valor, entretanto não diferiu da silagem NI, $LPP_1$ e $LPP_2$. A fração "c" da MS e MO não diferiram entre os aditivos. Conclui-se que a silagem com IGCG

apresentou o melhor perfil fermentativo e o melhor valor nutritivo, proporcionando aumentos na degradabilidade ruminal da MS, MO do Capiaçu, melhorando a degradação dos nutrientes, sendo uma alternativa para a maior disponibilidade de nutrientes no rúmen.

**Palavras-chave**: Aditivos. Degradabilidade ruminal *in situ.* Perfil fermentativo. Silagem de BRS Capiaçu. Valor nutricional.

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**SUMMARY**

## INTERPRETATIVE SUMMARY

**Effects of additives on fermentation profile and *in situ* rumen degradability of elephant grass silage cv. BRS Capiaçu.** By Santos et al. Elephant grass cv. BRS Capiaçu has high productivity and can be used for dairy cattle as silage. However, if an additive is not used, low-quality silage may be produced, compromising animal production. The objective of this study was to evaluate the effects of four doses of different bacterial additives or the inclusion of 8% ground corn grain in Capiaçu silage on the fermentative profile, chemical composition and rumen degradability *in situ*. Silage with 8% ground corn grain showed the best fermentative profile, nutritional value, and increased rumen degradability of dry matter and organic matter, improving nutrient degradation.

**Running title: Effects of additives on the fermentation profile and ruminal degradability of Capiaçu silage.**

Luciano S. Santos[1], Alex L. Silva[1], Bernardo M. Martins[1], Dayanne L. Sousa[1], Kellen R. Oliveira[1], Odilon G. Pereira[1], João V.C. Rodrigues[1], Poliana T.R. Salgado[1], Luis H. Silva[1], Gabriel Fernandes[2], and Polyana P. Rotta[1*]

[1] *Department of Animal Science, Universidade Federal de Viçosa, Viçosa, 36570-900, Brazil.*

[2] *Department of Agronomy, Universidade Federal de Viçosa, Viçosa, 36570-900, Brazil.*

* Corresponding author: Polyana Pizzi Rotta. Mailing address: Av. PH Rolfs – Campus UFV. 36570-900. Viçosa/MG, Brazil. E-mail: polyana.rotta@ufv.br, phone: +55 (31) 99449-1703.

**ABSTRACT**

The objective of this work was to evaluate the effects of four doses of different bacterial additives or the inclusion of 8% ground corn grain in elephant grass cv. BRS Capiaçu on the silage fermentative profile, chemical composition, and *in situ* ruminal degradability. The experimental design was completely randomized, with treatments set up in a 6 × 3 factorial scheme (six additives × three fermentation periods) and three replications. The additives were 1) no inoculant (NI); 2) 0.5 g/ton of commercial product composed of *Lactobacillus buchneri* ($LBU_{0.5}$); 3) 1 g/ton of commercial product composed of *Lactobacillus buchneri* ($LBU_1$); 4) 1 g/ton of commercial product composed of *Lactobacillus plantarum* and *Pediococcus acidilactici* ($LPP_1$); 5) 2 g/ton of commercial product composed of *Lactobacillus plantarum* and *Pediococcus acidilactici* ($LPP_2$); and 6) inclusion of 8% ground corn grain (IGCG) in the silage, which was opened at 30, 60 and 90 d of fermentation. The silage opened at 60 d of fermentation was subjected to *in situ* rumen degradability procedures using three lactating Holstein cows fistulated in the rumen, allowing for the following incubation times (0, 3, 6, 12, 24, 30, 48, 72, 120 and 240 h). Silage with IGCG showed the highest levels of dry matter (DM), crude protein, and other extract and reductions in the neutral detergent fiber (NDF), acid detergent fiber and lignin. For the fermentation profile, silages containing IGCG showed the lowest pH, butyric acid, and ammoniacal nitrogen levels and the highest lactic acid levels. Water-soluble carbohydrates and total DM losses did not differ between treatments. Silages containing IGCG showed greater potential and effective degradability of DM and organic matter (OM), with a consequent increase in fraction "a" (soluble fraction) and a reduction in fraction "b" (potentially degradable insoluble fraction). The potential degradability of NDF showed the highest values, however, differing only from silages supplemented with $LBU_1$. The effective degradability of NDF and its fractions "C" (the degradation rate of the potentially degradable fraction) and "I" (the indigestible fraction) was not influenced by the use of additives. When the "D" fraction (the potentially degradable fraction of the NDF) was

observed, the silage containing IGCG presented the highest value; however, it did not differ from the NI, $LPP_1$ and $LPP_2$ silages. DM and MO fraction "c" did not differ between additives. It is concluded that the silage containing IGCG presented the best fermentative profile and the best nutritional value, providing increases in the ruminal degradability of DM and Capiaçu MO, improving the degradation of nutrients, and providing an alternative for the availability of nutrients in the rumen.

**Key words:** Additives, BRS Capiaçu silage, fermentation profile, nutritional value, *in situ* ruminal degradability.

## INTRODUCTION

Approximately 23.5% of Brazilian dairy farmers use tropical grasses for silage production, primarily elephant grass (*Pennisetum purpureum,* Schum.; Bernardes and Rêgo, 2014). This genus has a high potential for biomass production, easy adaptation to different ecosystems, good nutrient composition if well managed and, above all, good acceptability by dairy cattle (Muck et al., 2018). Moreover, it is a good grass silage alternative for tropical countries around the world.

In 2016, a new cultivar of elephant grass cv. BRS Capiaçu was launched, and it is characterized as the most productive cultivar in the genus (50 tons of DM/ha/yr.; Pereira et al., 2017). This grass may be used to feed dairy herds in the forms of chopped feed and/or silage (Pereira et al., 2016). However, the ensiled form is more attractive to dairy producers due to the lower need for daily labor.

The possibility of implementing elephant grass cv. BRS Capiaçu on dairy farms where the cultivation of corn for silage production is unfeasible, due to water deficiency and/or limited area, is recommended. A major issue in producing elephant grass silage (*Pennisetum purpureum,* Schum.*)*, including the new cultivar BRS Capiaçu, is that it has a high moisture content, low soluble carbohydrate content, and high buffering capacity at ensiling time (Ferreira et al., 2013). Due to these characteristics, there is a slow reduction in pH, which provides conditions for undesirable bacterial development, such as *Bacillus*, *Listeria*, and *Clostridium*, producing low-quality silage to feed to dairy cattle (Amaral et al., 2020). Therefore, to produce high-quality elephant grass silage, the use of additives is essential, whether they are additives such as bacterial inoculants or moisture sequestering additives (Borreani et al., 2018, Muck et al., 2018).

The addition of additives containing homofermentative lactic acid bacteria (*Lactobacillus plantarum* and *Pediococcus acidilactici*) is extremely important, because the correct use of

these additives aimed at preventing losses of nutrients through the generation of $CO_2$ by undesirable fermentations (Santos and Zanine, 2006). The use of additives with heterofermentative bacteria, such as *Lactobacillus buchneri*, is intended to metabolize lactic acid and glucose during fermentation, synthesizing acetic and propionic acids that are effective in controlling fungi and other spoilage microorganisms and improving the stability of the fermentation (Kleinschmit and Kung, 2006).

Adding an absorbent to silage should reduce losses during the fermentation process (Andriguetto et al., 2003). In this context, ground corn grain may be an effective additive for reducing gas losses, effluent losses, and DM losses and increasing the digestibility of fibrous fractions, improving the nutritional silage value (Paula et al., 2020).

The literature has little information on the use of this cultivar for silage production with additives containing homofermentative and heterofermentative lactic acid bacteria and the addition of ground corn grain. In view of this scarcity, it is essential to develop studies that establish technical recommendations with doses of additives for their use as silage, ensuring alternative feed of excellent quality for dairy cattle.

Our hypothesis is that using 8% ground corn grain in elephant grass silage cv. Capiaçu will reduce fermentative losses and improve the nutritional value, with a consequent modification of the chemical composition, which would provide better ruminal degradability of the DM, OM and NDF fractions of the silage.

Thus, the objective of the present study was to evaluate the effects of four doses of different bacterial additives or the addition of 8% ground corn grain to Capiaçu silage on the silage fermentative profile, chemical composition, and ruminal degradability *in situ*.

## MATERIAL AND METHODS

### *Study Location and Experimental Design*

The experimental design used here was completely randomized, with the treatments set up in a 6 × 3 factorial scheme (six additives × three fermentation periods) and three replicates. The additives were: 1) no inoculant (**NI**), 2) 0.5 g/ton of a commercial product composed of *Lactobacillus buchneri* (**$LBU_{0.5}$**), with minimum guarantee levels of $1.0 \times 10^{11}$ CFU/g, at a final inoculation rate of $5.0 \times 10^{4}$ CFU/g of forage; 3) 1 g/ton of a commercial product composed of *Lactobacillus buchneri* (**$LBU_1$**), with minimum guarantee levels of $1.0 \times 10^{11}$ CFU/g, at a final inoculation rate of $1.0 \times 10^{5}$ CFU/g of forage; 4); 1 g/ton of a commercial product composed of *Lactobacillus plantarum* (**$LPP_1$**) with minimum guarantee levels of $5.0 \times 10^{10}$ CFU/g and *Pediococcus acidilactici*, with minimum guarantee levels of $1.5 \times 10^{10}$ CFU/g, reaching a final inoculation rate of $6.5 \times 10^{4}$ CFU/g of forage; 5) 2 g/ton; of a commercial product composed of *Lactobacillus plantarum* (**$LPP_2$**), with minimum guarantee levels of $5.0 \times 10^{10}$ CFU/g and *Pediococcus acidilactici*, with minimum guarantee levels of $1.5 \times 10^{10}$ CFU/g, at a final inoculation rate of 1.3 × 105 CFU/g of forage; and 6) 8% ground corn grain (**IGCG**). The additives described above were applied to the forage mass and later ensiled in mini silos that were opened at 30, 60, and 90 d of fermentation. The chemical composition of elephant grass before ensiling is shown in Table 1.

### *Planting and Harvesting Management*

The soil was conventionally prepared using plowing and harrowing over a total area of 0.7 ha. The planting was performed at the beginning of the rainy season in Brazil on October 29,

2020, with furrows measuring approximately 25 cm in depth spaced to provide 1.30 m between rows, and the crop was planted in vegetative form with double seedlings in the tip-with-foot system. The number of seedlings used for the study was 6 ton/ha.

Establishment fertilization was based on the results of a soil analysis, by using only phosphate fertilizer (single superphosphate) at planting, which was distributed at the bottom of the furrows. A total of 100 kg/ha of $P_2O_5$ was used, which corresponds to 500 kg/ha of single superphosphate. The first topdressing fertilization was performed when the plants reached an average height of 50 cm with an NPK formulation (25-00-25) using 87.5 kg/ha of N and 87.5 kg/ha of $K_2O$, which corresponds to 350 kg/ha of 25-00-25 fertilizer.

One day before harvest, 100 plants were sampled from an area measuring 0.7 ha and taken to the laboratory where the average height of the grass was measured, and then the leaf blade, stem and dead matter were separated from each plant and the dead material was weighed, then the proportions of leaves, stem, dead matter, and the total productivity per hectare (kg/ha) based on the DM were estimated to evaluate the quality of the material that was ensiled.

The elephant grass harvest cv. Capiaçu was performed on February 26, 2021, and it presented approximately 14% DM after 120 d of growth. For harvesting, a mechanical harvester was used (Modelo PREMIUM FLEX 2008, Menta Mit, São Paulo, Brazil), and the forage was cut into a theoretical cutting size of 11 mm.

***Production of Elephant Grass Silage cv. BRS Capiaçu***

After the forage was cut in the field, 15 kg of fresh forage was separated for each mini silo, and then the bacterial additives were applied to the 15 kg forage mass separately for the following treatments: $LBU_{0.5}$, $LBU_1$, $LPP_1$, and $LPP_2$. To another 15 kg of fresh forage, 8%

ground corn grain was added. The bacterial additives were mixed with pure distilled water and sprayed on the forage; each silo was sprayed individually. The same volume of pure distilled water was added to the NI treatment.

The silage was maintained in experimental silos consisting of plastic buckets with a sealing lid capacity of 10 L, a height of 29.5 cm, an upper diameter of 24 cm, and a base diameter of 22 cm. A total of 3 kg of sand and a cloth were placed at the bottom of each bucket to separate the silage from the sand, in which the effluent was retained. Fifty-four experimental silos were produced, representing 3 replications for each treatment. The forage was compacted with wooden sockets. At the end of the ensiling process, the experimental silos were weighed, sealed with a bucket lid and adhesive tape for later isolation and stored in a covered place protected from the sun and rain. The mini silos were opened after 30, 60, and 90 d of fermentation.

***Fermentation Profile of Elephant Grass Silages cv. BRS Capiaçu***

When the mini silos were opened, the top 10 cm of Capiaçu silage was discarded from each experimental silo, and the other part of the silage was homogenized and sampled for further chemical analysis and fermentation profiling. An aqueous extract was prepared from 25 g of silage sample and 225 mL of distilled water homogenized in a blender for 1 min. The aqueous extract was used to evaluate the pH using a digital potentiometer (Tecnal, model Tec-3MP), acidified 1:1 with $H_2SO_4$, and diluted with distilled water for further analysis of the $NH_3$-N using the phenol-hypochlorite method (Okuda, 1965). Soluble carbohydrates were analyzed as described by Nelson (1944), and organic acids (lactic, acetic, propionic, and butyric acids) were analyzed according to the methodology described by Siegrifield et al. (1984).

Samples used for organic acid quantification were treated with calcium hydroxide and cupric sulfate and analyzed using HPLC (SPD-10 AVP, *Shimadzu*, OR, USA) according to the method by Siegrifield et al. (1984). The HPLC apparatus (SPD-10 AVP, Shimadzu) was equipped with a refractive index detector and an Aminex HPX-87H column (BIO-RAD, CA, United States), with the mobile phase containing 0.005 M $H_2SO_4$ and a flow rate of 0.7 mL/min at 45 °C.

For buffer capacity estimation, the methodology by Playne and McDonald (1966) was used, which involved the maceration of approximately 15 g of fresh forage and its dilution in 250 mL of distilled water. Then, titration was performed to pH 3.0 with HCl (0.1 N) and the samples were subsequently titrated with NaOH (0.1 N) to pH 6.

***Chemical Composition***

Samples of the ensiled material were dried at 55 °C in a forced air oven for 72 h and then ground in a 1 mm sieve in a Wiley mill (Fortinox, model STAR FT 50/6) and subsequently analyzed for DM, (Method INCT-CA G-003/1), ash (Method INCT-CA M-001/2), CP (Method INCT-CA N-001/2), ether extract (EE, INCT-CA Method G-005/2), NDF (INCT-CA Method F-002/2), ADF (INCT-CA Method F-004/2), lignin (INCT-CA Method F-005/2), hemicellulose (INCT-CA Method F-005/2) and acid detergent insoluble protein (ADIP, INCT-CA Method N-005/1) according to Detmann et al. (2021). Nonfibrous carbohydrates were calculated as 100 - (NDF + CP + EE + ash; Hall, 1999).

***Total Dry Matter Losses and Losses from Gases and Effluents***

These measurements were obtained by weighing the empty and full laboratory silos before and after ensiling and weighing their respective DM contents, according to the methodologies established by Jobim et al. (2007).

The determination of total DM losses was calculated by finding the difference between the initial and final gross weights of DM in the silos in relation to the amount of forage mass that was ensiled according to the following equation described by Schmidt (2006):

$$TLDM = \frac{[(ADMi - ADMf)]}{ADMi} \times 100$$

where:

TLDM = total loss of DM (% DM)

ADMi = amount of initial DM. Weight of the silo after filling - weight of the empty set, without forage before filling × DM content of the forage in the silage.

ADMf = amount of final DM. Weight of the filled silo before opening - weight of the empty set, without the forage, after opening the silos × forage DM content at opening.

The measurement of gas losses during the silage process was obtained by weighing the laboratory silos at closing and opening times in relation to the stored forage mass. The following equation was used to estimate the gas losses according to Mari (2003):

$$GL = \frac{(\mathrm{SWs} - SWo)}{(FMs \times \mathrm{DMFs})} \times 100$$

where:

GL = gas losses during storage (% of initial DM);

$SW_S$ = silo weight at silage (kg);

$SW_O$ = silo weight at opening (kg);

$FM_S$= forage mass at silage (kg);

DMFs = DM content of forage in silage (kg)

Effluent production was calculated through the strategic use of dry sand at the bottom of the laboratory silos. The effluent production was determined by finding the difference in the weight of the silo + sand + cover + cloth at opening (after removing the forage from the silo) and before ensiling in relation to the amount of fresh forage that was ensiled. The following equation was used according to Schmidt (2006):

$$EP = \frac{(WTSo - WTSi)}{(GEFm)} \times 1000$$

where:

EP = effluent production (kg/ton of as fed mass);

$WTS_o$ = weight of the set (silo + sand + cloth + lid) at opening (kg);

$WTS_i$ = weight of the set (silo + sand + cloth + cover) at the silage process (kg);

$GEF_m$ = green ensiled forage mass (kg)

### *In Situ* Ruminal Degradability Procedures

All procedures involving the use of animals were previously submitted to the Ethics Committee on the Use of Research Animals of the Federal University of Viçosa and were approved under protocol number 60/2022.

The silages that were opened at 60 d of fermentation were analyzed for *in situ* rumen degradability.

### *Animals, Facilities and Experimental Design*

The experiment was performed at the Dairy Unit of the Teaching and Research Farm at the Animal Science Department of the Universidade Federal de Viçosa. Three lactating Holstein cows cannulated in the rumen with characteristics of being 5 ± 1.3 yr. old, having 650 ± 50.7 kg BW, and 30 ± 5.2 kg/d of milk production were used. The cows were distributed in a randomized block design, in which the three animals represented the blocks. Thus, we had 6 treatments (6 additives per mini silos × 1 fermentation period of 60 d) and 3 replications (animals) during each period. Each treatment group was incubated in the rumen of a different cow.

The cows were kept in a free stall system, with a concrete floor, sand beds, feeder, and water ad libitum. They were adapted for 14 d (Machado et al., 2016) with corn silage and concentrate (Table 2). The same roughage:concentrate ratio (55:45) was maintained throughout the evaluation period of *in situ* ruminal degradability. After the adaptation period, rumen degradability procedures were performed, which lasted for 10 d of evaluation.

***In situ Degradation Procedures***

After the mini silos that had reached 60 d of fermentation were opened, the top 10 cm of the Capiaçu silage was discarded from each experimental silo. The remaining silage was homogenized and sampled at -20 °C. After being thawed, proportional (600 g) samples of each repetition were mixed to obtain a composite sample of each additive silage and later placed at 55 °C for 72 h. Later, the samples were ground using a Wiley mill (Fortinox, model STAR FT 50/6) at a 2 mm grain size.

Approximately 4 g of each dry sample was individually weighed into nylon bags (Sefar Nitex, Switzerland; porosity 50 μm, 10 × 20 cm) and rumen-incubated. The samples were

placed in individual bags at a ratio of 20 mg DM/$cm^2$ of bag surface area (Nocek, 1988). Incubation was performed for the following ruminal degradation times: 0, 3, 6, 12, 24, 30, 48, 72, 120, and 240 h (Detmann et al., 2021). The number of nylon bags varied as a function of the incubation time to ensure sufficient residual samples after incubation (i.e., more bags per sample were incubated for longer incubation times).

The samples were incubated in the rumen by attaching the nylon bags to a steel chain (90 × 2 $cm^2$; Menezes et al., 2019) with a weight (300 g) at the end to allow for continuous immersion in the rumen contents. The bags were placed in the rumen in the reverse order of the incubation hours, so all the bags were removed at the same time and washed. After the end of incubation, the bags were washed in running cold water. The end point to finish the washing bags was the high clarity of the rinse water (Zanetti et al., 2017).

The bags were dried in a forced ventilation oven at 55 °C for 72 h and then placed in an oven at 105 °C for 2 h to estimate the disappearance of rumen DM (Menezes et al., 2019; Silva et al., 2019). The 0 h nylon bags were immersed in the rumen according to the methodology by Detmann et al. (2021), and later, they were rinsed in running water and subjected to the same procedures mentioned above.

The residue of each sample per treatment (silage with additives that had reached 60 d of fermentation) was removed from the nylon bags, ground in a Wiley mill (Fortinox, model STAR FT 50/6) with a 1 mm sieve, and placed in a labeled plastic bag to obtain a sample from each animal and its respective incubation/treatment time. Residual samples were analyzed for DM, OM, and NDF according to the methodology described by Detmann et al. (2021) and were later used to estimate the ruminal degradation parameters of DM, OM, and NDF.

***Degradation Models***

The *in situ* degradability of DM and OM was estimated by finding the weight difference of each component between the weighing before and after ruminal incubation and was expressed in g/kg DM. The coefficients "a", "b", and "c" of the DM and OM were found according to Ørskov and McDonald (1979) using the following equation:

$$Deg(t) = a + b\ (1 - \ e^{(-ct)})$$

where Deg (t)= represents the degradability or disappearance of the constituent (DM, OM) of the feed, as expressed in g/kg of DM; a = is the water-soluble fraction of the feed at time zero; b = is the fraction that is insoluble in water but is potentially degradable in the rumen at a given time; c = is the degradation rate of the potentially degradable fraction in the rumen (b); and t = is the incubation time (h).

The *in situ* degradability of NDF was also estimated by finding the weight difference between the weighings performed before and after ruminal incubation and was expressed in g/kg of DM. To obtain the NDF "D", "C". and "I" coefficients, the model proposed by Waldo et al. (1972) was used:

$$R(t) = D(e^{-ct}) + I$$

where R(t)= is the incubation residue at time t (h); D= is the fraction of potentially degradable NDF in the rumen; C= is the rate of degradation of fraction D; and I= is the nondegradable fraction of NDF.

The effective degradability (ED) of DM and OM in the rumen was calculated using the following model:

$$ED = A + \ (B\ \times c\ /\ c + \ K)$$

where k corresponds to the estimated rate of passage by particles in the rumen, which were used at 2, 5 and 8 $h^{-1}$, simulating low, medium and high passage rates, respectively.

To calculate the effective degradability of NDF, the following model was used:

$$ED = BP \times c/(c + k)$$

where BP is the standardized potentially degradable fraction (%).

***Statistical Analyses***

The data were analyzed in a completely randomized design in a 6 × 3 factorial arrangement. The model includes the fixed effect of additives, the fermentation period and the interaction between these factors, according to the following statistical model:

$$Yijk = \mu + \alpha i + \beta j + (\alpha\beta)ij + \varepsilon ijk,$$

where Yijk = dependent variable, μ = general average, αi = fixed effect of the i-th additive, βj = fixed effect of days of fermentation, (αβ) ij = interaction between additives and days of fermentation, and εijk = random error.

The nonlinear models for each treatment and time of fermentation were adjusted by the cow level using the nls function in R (R Development Core Team, 2015) for each animal and treatment. Subsequently, the extracted function parameters were organized in a dataset format including the effects of the treatments and fermentation time.

R software was used for the analysis of variance and comparison of means. Means were separated by Tukey's test, and differences were declared at the level of $P < 0.05$.

# RESULTS

## *Agronomic Characterization and Buffering Capacity of BRS Capiaçu Elephant Grass*

The BRS Capiaçu elephant grass was sampled 1 d before harvest, and it had an average height of 3.60 m and the following yields: 65.8% culms, 28.2% leaves, and 6.02% dead matter with an estimated total productivity of 15.843 kg/ha DM. The buffer capacity of BRS Capiaçu for different treatments is presented in Table 1.

## *Chemical Composition of Silages*

There was no interaction ($P > 0.05$) between the additives and fermentation days for all the variables related to chemical composition. However, the DM content varied ($P < 0.001$) according to the fermentation days, presenting the lowest contents at 60 and 90 d compared to 30 d of fermentation (Table 3). In relation to CP, we observed that 30 d of fermentation presented a greater ($P < 0.001$) value in comparison with 60 and 90 d.

The neutral detergent fiber and ADF contents were influenced ($P < 0.001$) by fermentation days (Table 3). The NDF levels were similar at 30 and 60 d of fermentation, but at 90 d of fermentation, a greater ($P < 0.001$) NDF value was observed. Regarding the ADF content, we observed that as the fermentation days increased, the content also increased differently among fermentation days. The greatest ($P < 0.001$) DM and CP levels were observed for IGCG compared to $LBU_{0.5}$, $LBU_1$, $LPP_1$, $LPP_2$ and NI.

The NDF and ADF levels were influenced ($P < 0.001$) by the additives used here, with the lowest values for silage with IGCG. The silage additive with IGCG showed averages of 494 and 327 g/kg DM for NDF and ADF, respectively, differing from the values of silages with

bacterial additives and NI with averages of 687 and 700 g/kg of DM for NDF and 472 and 480 g/kg from DM to ADF, respectively. The lignin and hemicellulose contents also displayed considerable reductions, with the IGCG silage presenting averages of 60.0 and 167 g/kg DM, respectively, differing from the values of silages containing bacterial additives (Table 3).

***Fermentation Profile of Silages***

There was no interaction ($P > 0.05$) between additives and fermentation days (Table 4). However, the pH, water soluble carbohydrates (**WSC**), and total DM loss contents differed ($P < 0.001$) among fermentation days, with greater values at 60 and 90 d (Table 4). The total DM losses were approximately 275% when comparing 30 d with 60 and 90 d of fermentation. The lactic/acetic acid ratios decreased over the fermentation days, and at 30 d of fermentation, the silage presented a proportion of 2.64 g/kg DM. At fermentation days 60 and 90, the content decreased, stabilizing at 60 d, with values of 0.51 g/kg DM. Regarding effluent production (EP), there was no effect ($P > 0.05$) of fermentation days (Table 4).

There was an effect ($P < 0.05$) of the additives on the pH, EP, and lactic/acetic acid ratio, with lower pH values for silage with IGCG, differing from NI and additives. Regarding EP, the $LPP_2$ treatment had the greatest value, but it did not differ from the $LBU_{0.5}$, $LBU_1$, and $LPP_1$ (Table 4). For the effects of the additives, the silage containing IGCG had the greatest proportion of lactic/acetic acid, differing from NI and bacterial additive silages with averages of 3.375, 0.832, and 0.776 g/kg, respectively.

An interaction ($P < 0.001$) was observed between additives and fermentation days for ammonia nitrogen (TN) and gas losses (GL) (Table 5). The lowest TN value ($P < 0.001$) was

observed for silages supplemented with IGCG and $LBU_{0.5}$ at 60 d of fermentation (Table 5). At 90 d of fermentation, a lower ($P < 0.001$) concentration of TN for IGCG was observed, differing in relation to bacterial additives and NI (Table 5). At 30 d of fermentation, there was no significant difference between all additives and NI. The TN concentration did not differ between the opening times of 30, 60 and 90 d for the silage supplemented with IGCG. For the silages supplemented with LBU0.5 and LBU1, there was an increase in the concentration of this metabolite as the fermentation time increased. The same behavior was observed for silages supplemented with LPP1, LPP2 and NI, with increases in the TN content from 30 to 90 d and stabilization at 60 d of fermentation (Table 5).

Regarding gas losses (Table 5), although the silage supplemented with IGCG showed the lowest GL at 30 and 60 d of fermentation, it did not differ from NI and silage supplemented with bacterial additives (Table 5). At 90 d of fermentation, a higher GL was observed for NI silages, differing from all the additives, which did not differ between them, although the silage with IGCG had a lower numerical value, which was not statistically significant (Table 5). The GL concentration did not differ between the fermentation periods of 30, 60 and 90 d for the silage supplemented with IGCG. For NI silages, there was an increase in the concentration of this metabolite as the fermentation time increased (Table 5). In the silages supplemented with $LBU_{0.5}$ and $LBU_1$, there were increases in the GL from the fermentation period of 30 to 60 d, when it stabilized. The silages supplemented with $LPP_1$ and $LPP_2$ showed an increase in losses, stabilizing at 60 days of fermentation (Table 5).

***Profile of Organic Acids in Elephant Grass Silages BRS Capiaçu***

An interaction (P < 0.001) was observed between additives and fermentation days for all organic acids evaluated here (Table 5). A greater lactic acid (P < 0.001) value was observed for silage additives $LBU_{0.5}$, $LPP_{1,}$ and IGCG at 30 d of fermentation (Table 5). At 60 and 90 d of fermentation, a greater (*P* < 0.001) concentration of lactic acid was observed for IGCG, differing in relation to the doses of bacterial additives and NI. The concentration of lactic acid was greater at 30 d, differing from 60 and 90 d of fermentation for all treatments (additives) evaluated here.

In relation to acetic acid, we observed that at 30 d of fermentation, the treatment containing IGCG presented a lower value (P < 0.001) in relation to the $LBU_1$ treatment; however, these treatments did not differ from NI, $LBU_{0.5}$, $LPP_1$, and $LPP_2$ (Table 5). At 60 d of fermentation, a lower content (P <0.001) was observed for the treatment containing IGCG, followed by $LBU_1$, with averages of 5.26 and 15.64 g/kg DM, respectively. At 90 d of fermentation, the silage supplemented with IGCG showed the lowest (*P* < 0.001) concentration of acetic acid, differing from the NI, $LBU_{0.5}$, $LBU_1$, $LPP_1$, and $LPP_2$ treatments, which differed only from $LBU_{0.5}$ and $LBU_1$. Throughout the evaluation, we observed that the acetic acid content when using $LBU_{0.5}$, $LBU_1$, $LPP_1$, and $LPP_2$ increased from 30 to 60 d of fermentation, and when observing the results at 90 d, the acetic acid was reduced again, differing from 60 d with the exception of $LBU_1$, which was similar between 60 and 90 d. The same trend was not observed for acetic acid NI, which increased and stabilized at 60 d of fermentation. The silage additive containing IGCG showed no difference in acetic acid over the fermentation days, with an average of 4.77 g/kg DM (Table 5).

For NI and $LBU_{0.5}$ silages, we observed differences (*P* < 0.001) in propionic acid at 30, 60, and 90 d of fermentation (Table 5). However, for IGCG silage, the propionic acid was similar among fermentation days. The $LBU_1$, $LPP_1$, and $LPP_2$ silages presented greater (P < 0.001) propionic acid at 60 and 90 d of fermentation in relation to 30 d (Table 5).

With the exception of LPP1, the butyric acid increased ($P < 0.001$) for all additives between 30 and 60 d of fermentation (Table 5). For the NI and $LPP_2$ silages, butyric acid did not change ($P > 0.05$) between 60 and 90 d. However, for the $LBU_{0.5}$ and $LPP_1$ silages, butyric acid was greater at 90 d than at 60 d. Similarly, for the $LBU_1$ and IGCG silages, we observed a decrease in butyric acid at 90 d in comparison to 60 d (Table 5). At 90 d of fermentation, the silage supplemented with IGCG had the lowest butyric acid content, differing from the silage supplemented with bacterial additives and NI, with an average of 21.2 g/kg DM (Table 5). At 30 d of fermentation, the same behavior was observed for silage containing IGCG; however, it did not differ from silages supplemented with $LBU_{0.5}$ and $LPP_2$, with means of 12.1, 15.0 and 11.2, respectively.

**In Situ Ruminal Degradability Parameters of Elephant Grass cv. BRS Capiaçu**

There was an effect of the additives ($P < 0.001$) for fraction “a” of the DM and OM, with considerable increases for the silage containing IGCG and differing from the bacterial and NI additives, presenting values of 419 and 400 g/kg of DM, respectively, for additive silages with IGCG (Table 6).

Effects from additives ($P < 0.001$) were observed for the digestibility fraction “b” of DM and OM (Table 6). We observed that fraction "b" was reduced for silages supplemented with IGCG, differing from silages with bacterial additives and NI. Regarding the use of bacterial additives and NI, there was no substantial variation in the values of this fraction (Table 6).

Despite the effects of additives on fractions “a” and “b” of DM and OM, there was no effect ($P > 0.05$) between additives for fraction “c” of DM and OM (Table 6). The DM and OM degradation rates in the $\%/h^{-1}$ (fraction "c") remained constant, with general averages of 0.0202 and $0.0205\%/h^{-1}$, respectively (Table 6).

In NDF, there was an effect ($P < 0.05$) of the additives on its "D" fraction, with the silages supplemented with IGCG presenting the highest values (578 g/kg of DM), not differing only from NI and LPP1, LPP2 silages (Table 6). However, NI silages and those supplemented with bacterial additives did not differ statistically from one another (Table 6).

In the absence of inoculation, the levels of bacterial additives and IGCG did not change the "C" and "I" fractions of the NDF ($P \geq 0.05$), with general averages of 0.019%/$h^{-1}$ and 376.5 g/kg of MS (Table 6).

There was an effect of additives ($P < 0.001$) on the potential degradability of DM and OM (Table 7). With the silages supplemented with IGCG, the highest values of 780 and 770 g/kg DM were observed, respectively. The lowest values for the potential degradability of DM and OM were observed for NI silage and with bacterial additives, which did not differ from each other, with general averages of 675, 682 and 653 and 658 g/kg DM, respectively. (Table 7).

The same trend observed for potential degradability was observed for the effective degradability of MS and MO (Table 8). The highest values ($P < 0.001$) were observed for silage with IGCG when compared to NI silage and with bacterial additives (Table 8). When considering all passage rates (2, 5 and 8%/$h^{-1}$) for IGCG silages, except for the potential degradability of OM at the passage rate of 8%/$h^{-1}$, which was approximately 487 g/kg DM, all the other silages with IGCG showed effective DM and OM degradability above 500 g/kg DM (Table 8).

The potential degradability of NDF of silages supplemented with IGCG showed higher values ($P < 0.05$); however, this degradability did not differ compared with treatments NI and $LBU_{0.5}$, $LPP_1$ and $LPP_2$. The lowest values were observed for silage supplemented with

$LBU_1$, which did not differ statistically from the NI treatments and bacterial additives (Table 7).

There was no effect of additives ($P > 0.05$) on the effective degradability of NDF for all passage rates evaluated here (2, 5 and 8%/per $h^{-1}$), with averages of 287, 162 and 113 g/kg DM per $h^{-1}$, respectively (Table 8).

We observed higher DM and OM degradability for IGCG silage when compared to NI and with bacterial additives for all incubation times (Fig. 1, 2). The maximum potential for DM and OM degradation of all silages was obtained after 240 h of incubation.

After 30 h of ruminal incubation, the DM and OM degradation values of IGCG silage were high (above 650 g/kg DM; Figs. 1, 2). No stabilization of OM and DM degradability was observed for the different treatments, showing that the silages still had the potential to be degraded after 240 h (Fig. 1, 2). It is important to emphasize that after 120 h of incubation, there is a deceleration in the degradation of silages in the rumen.

The NDF degradation curve of the silages shows that they differed in terms of the fiber degradation rate. Until the first 24 h of ruminal incubation for silages, NDF degradation remained close to 289 g/kg DM (Fig. 3). The IGCG silage reached 240 h of incubation with an average of 653 g/kg DM.

However, for silages spiked with IGCG, the degradation of NDF after 24 h of incubation was higher when compared to NI silages and silages spiked with bacterial additives (Fig. 3). We observed that the degradability of NDF after 120 h of ruminal incubation presented the highest value (above 500 g/kg of DM). However, no stabilization in the degradability of the silages was observed, showing that the silages still showed degradability potential after 240 h (Fig. 3). However, there is a notable deceleration after 120 h of ruminal incubation.

## DISCUSSION

The elephant grass BRS Capiaçu was harvested at a height of 3.60 m and presented a total productivity of 15.843 kg/ha of DM and the following characteristics of the plant fractions: 65.8% stems, 28.2% leaves, and 6.02% dead matter based on DM. The agronomic characteristics of BRS Capiaçu elephant grass sampled one day before harvest were similar to the values found in the literature by Pereira et al. (2017), Alves (2021), and Retore et al. (2021). However, the DM content before ensiling, which ranged from 133.2 to 192.4 g/kg DM, was below 300 g/kg DM, a level considered adequate for good mass fermentation as well as to eliminate or minimize the effects of effluent production (McDonald et al., 1991), which may partly explain the predominant butyric fermentation in our study.

The buffer capacity of the elephant grass plant BRS Capiaçu with IGCG in relation to NI, $LBU_{0.5}$, $LBU_1$, $LPP_1$, and $LPP_2$ was numerically lower. However, silage containing IGCG resulted in a greater reduction in pH after the ensiling process in relation to doses of bacterial additives and NI, which may be due to the lower resistance of the plant to inhibit the pH drop after ensiling (Rodrigues et al., 2005). The silage containing IGCG presented a lower pH because of the addition of ground corn grain in grass silage, which was essential to reducing its buffer capacity and helped to provide a lower post-silage pH, presenting pH values within the ideal for BRS Capiaçu elephant grass silage (Amaral et al., 2020; Paula et al., 2020; Ribas et al., 2021).

A high concentration of lactic acid was observed for silages containing IGCG when compared to NI and doses of bacterial additives at 60 and 90 d of fermentation. This result is due to the addition of ground corn grain to elephant grass silage, which provided WSC and helped reduce the pH, because the lactic acid bacteria used this substrate for their growth, and there was a greater production of this metabolite (Widyastuti, 2008; Borreani et al., 2018).

The lactic acid contents in our study ranged from 12.7 to 19.5 g/kg DM between fermentation times of 30 and 90 d for IGCG silage, respectively, being slightly below other studies on elephant grass cv. BRS Capiaçu (Amaral et al., 2020; Jesus et al., 2021). However, the lactic acid concentration in the study conducted by Ribas et al. (2021) corroborates our results, allowing us to qualify silage with corn grain as satisfactory but NI and silage with bacterial additives as unsatisfactory. According to BREIREM and ULVESLI (1960), high-quality silage may have lactic acid levels of 15 to 25 g/kg DM.

The silages containing bacterial additives showed greater pH values, which is an indication of great butyric and acetic acid production, the result of unwanted fermentations (Kung Jr., 2018). The consequence of obtaining a silage with a high pH is that the development of clostridia may occur, and secondary fermentations may also occur, because it acts against the preservation of the silage by destroying the lactic acid, increasing the pH, and decreasing the nutritive value of the silage (McDonald et al., 1991). This finding was observed for silages inoculated with different bacterial additives and NI in our study.

The butyric acid content showed a lower value at 90 d of fermentation for silage with IGCG, differing from NI and bacterial additives in silages. This result is due to the increase in DM with the inclusion of ground corn grain, because low levels of DM in silages favor the growth of *Clostridium* bacteria, which is responsible for the production of this metabolite (Carvalho et al., 2015). The butyric acid concentrations for NI silage and added bacterial additives (with the exception of $LBU_{0.5}$ and $LPP_2$ at 30 and 60 d) at all fermentation times observed in this study were high, showing important clostridial fermentation. However, in this study, we did not have the bacterial population in the silages. The presence of this metabolite is correlated with undesirable fermentations, which can impair the quality of the final canned product (Lavezzo, 1985).

In observing the losses of gases and ammoniacal nitrogen at 90 d of fermentation, the NI treatment presented the greatest gas losses compared to silage containing bacterial additives and IGCG. For ammonia losses, there was a lower loss for silage with IGCG in relation to NI and bacterial inoculants at 60 and 90 d of fermentation. This finding may have occurred because gas losses result from secondary fermentation by *Clostridium*, enterobacteria, aerobic bacteria, and other microorganisms (McDonald, 1981; Muck, 1996). However, the losses of $NH_3$-N (TN%) were reduced for IGCG, which may have occurred because in silages with a lower pH, the bacteria that degrade proteins are inhibited, and thus proteolysis is reduced; consequently, the production of ammonia nitrogen is also reduced (Muck, 1996).

There was an effect of the additives on the DM and CP contents, with a significant increase in the levels of these nutrients with IGCG, differing from the NI and bacterial additives. The effect of increasing these nutrients for silage with IGCG may be due to the inclusion of ground corn grain acting as a moisture sequestrant and improving the nutritive value of the BRS Capiaçu elephant grass silage. The ground corn grain had approximately 88% DM and 9% CP with a good moisture retention capacity. When added to the inclusion level of 8% in Capiaçu silage, it ended up increasing the DM and CP contents, which is quite relevant in practical terms to produce a silage with very high quality.

In the very few studies found in the literature using ground corn grain as a moisture sequestrant in elephant grass silage cv. BRS Capiaçu, Paula et al. (2020) observed a linear increasing effect for DM and a decreasing effect for NDF and ADF contents. These authors stated that for each unit of ground corn grain added to silage, there was an increase of 0.94% in DM content. However, despite numerical increases in CP and ether extract contents in silages with inclusion ground corn grain, they did not differ from the control treatment.

By contrast, Andrade and Lavezzo (1998) tested different types of additives (wheat bran, corn husk, and saccharin) on the production of elephant grass cv. Napier silage and stated that

the silages that received inclusion levels of 16 and 24% of these additives had greater CP levels. Monteiro et al. (2011) observed increases in the CP content of silage supplemented with ground corn grain in relation to silages with no additive, sugar cane or bacterial inoculants.

The total DM losses were not influenced by the additives; however, the fermentation days influenced the total DM losses, with an increase of 275% between 30 and 90 d of fermentation, which is an indication of a quality reduction in the silage due to fermentation processes and high proteolysis (Junior and Lavezzo, 2001).

The amount of effluent produced in the silo is correlated with the moisture content of the ensiled material (Neto et al., 2021). Thus, decreasing the moisture content with the inclusion of IGCG is essential to reducing effluent losses in the silo and, consequently, DM losses. Paula et al. (2020) observed reductions in DM and effluent losses with the inclusion of increasing doses of ground corn grain. Thus, these authors recommend the inclusion of this additive in Capiaçu silages to reduce fermentative losses and to improve the nutritional value of the silage.

Andrade et al. (2012) observed that the use of 5% ground corn grain in elephant grass silage, with 15% DM, was effective in reducing effluent losses, not differing from the inclusion level of 10%, resulting in a reduction of 58%. The effluent losses observed in the present study were greater when compared to Andrade et al. (2012), who presented an average of 6.3 and 6.4 kg/$m^3$ for inclusion levels of 5 and 10%, respectively.

The elephant grass silage cv. BRS Capiaçu with IGCG showed the greatest values for fraction “a” of DM and OM, differing from NI and bacterial additives. This finding may be due to the contribution of ground corn grain addition in elephant grass silage with IGCG and because corn has a high value of this fraction in its composition (Carvalho et al., 2015). The

fraction “a” of DM for silage containing IGCG was 419 g/kg of DM, while for the bacterial additives, the NI was approximately 236 and 224 g/kg DM.

Ferreira et al. (2014) evaluated the *in situ* ruminal degradability of elephant grass silage with different bacterial additives and observed greater degradability for fraction "a" of DM for silage with *Streptococcus bovis* compared to silage with *Enterococcus* alone or no additive inclusion. These authors stated that the greater degradability of this fraction was due to the inclusion of *Streptococcus bovis* in the silage, which reduced the moisture content of the grass, contributing to the reduction of the pH and N-$NH_3$, favoring bacterial growth, increasing lactic acid production and reducing DM and CP losses. This result was also observed in our study on silage containing IGCG.

It is important to emphasize that fraction “a” of DM represents the portion of the silage that is readily available to rumen microorganisms (Costa et al., 2020). Therefore, our results for silage containing IGCG are very important for the use of nutrients at the rumen level. Carvalho et al. (2015) evaluated the ruminal degradability of elephant grass silage cv. Roxo and observed that with increasing levels of corn bran in the silage, the degradability of fraction “a” of DM increased linearly, yielding 315 g/kg DM with the inclusion level of 20 dag/kg of corn bran, and the silage using only the bacterial additives showed an average of 180 g/kg DM, which is very close to our results.

Negrão et al. (2014) evaluated the kinetic parameters of ruminal degradation for *Brachiaria decumbens* cv. Basilisk with different levels of rice bran inclusion, and fraction "a" of DM increased linearly. These authors stated that there was a linear increase of 0.54% for each 1% rice bran in the silage, yielding values for this fraction of 460 g/kg DM, with an inclusion level of 40% rice bran.

Despite the higher values for fraction "a" of DM and OM in IGCG, fraction “b” of silage (potentially degradable and insoluble) differed from the NI and bacterial additives. The lower values observed for fraction "b" for IGCG silage may be due to the low NIDA content of ground corn grain when compared to grass (2.69% of total-N). Insoluble nitrogen in acid detergent is the fraction of the feed that represents the N unavailable to the ruminal system (Carvalho et al., 2008), in which the inclusion level of 8% ground corn grain in elephant grass silage resulted in lower values of fraction "b" for DM and OM.

Pereira et al. (2000) studied the *in situ* ruminal degradability of elephant grass silage with increasing additions of corn and soybean processing residues, and they observed that fraction "b" linearly decreased with increasing inclusion levels, for values close to 43.4 and 53.2%, respectively, which corroborates the results observed in the present study.

It is clear that the use of bacterial additives in elephant grass did not cause great variation in the degradation of fractions "a" and "b" of DM and OM, because these bacterial additives did not provide any significant change in the chemical composition of our silages (Table 1), so they did not change their degradability. Ribas et al. (2021) evaluated the ruminal degradability of elephant grass silage cv. BRS Capiaçu, testing the effects of the drying times and the application of an enzymatic-bacterial additive mix (*Lactobacillus curvatus*, *L. acidophilus*, *L. plantarum*, *L. buchneri*, *L. lactis*, *Pediococcus acidilactici* and *Enterococcus faecium*). Ribas et al. (2021) observed no effect of the wilting time and additives on fractions “a” and “b” of DM in elephant grass silage. This finding was also observed in our results with or without bacterial additive inclusion for these fractions.

Despite the variations in fractions "a" and "b" observed in our studies, fraction "c" (DM and OM degradation rate in $\%/h^{-1}$) was not influenced by the additives, remaining constant with general averages of 0.0202 and $0.0205\%/h^{-1}$, respectively. These results are similar to the findings of Carvalho et al. (2015), who, in addition to having observed increases in fractions

"a" and "b" of DM for elephant grass silage with corn bran, stated that fraction "c" remained constant, with an average of 0.0220%/h[1].

Considering the NDF, the highest values of the "D" fraction (the potentially degradable fraction in the rumen) were observed in the silage containing IGCG; however, it did not differ from the silage with NI, $LPP_1$, and $LPP_2$. Silages supplemented with bacterial additives and NI did not differ from each other. This fact can be explained according to Carvalho et al. (2015), who states that the higher starch content present in ground corn grain compared to ensiled elephant grass may have provided a greater development of amylolytic bacteria in relation to cellulolytic bacteria, thus reducing the use of fiber, because it contained a material that was easier to use, which was ground corn kernels.

The use of additives in elephant grass silage did not influence the "C" (degradation rate of the "D" fraction) and "I" (nondegradable fraction of NDF) fractions of the NDF. All added silage did not differ between treatments. It is important to note that, in nutritional terms, for the ration to be considered high quality, the "C" fraction of the NDF must be between 0.02 and 0.06%/$h^{-1}$ (Mertens, 1993), and for all our evaluated silages, the average was 0.02%/$h^{-1}$.

Knowing the "D" and "I" NDF fractions is extremely important because through them, we may conclude that the "I" fraction (the nondegradable NDF fraction) has a relevant effect on the feed indigestibility (Cabral, 2002). Negrão et al. (2014) evaluated the ruminal degradability of NDF in grass silage and observed that the “C" fraction of NDF was not influenced by the inclusion of different levels of rice bran, with an average of approximately 0.051%/$h^{-1}$. However, there were considerable reductions in fraction "I" (indigestible fraction of the NDF) of grass with increasing rice bran levels.

The nonvariation in the NDF "I" fraction observed in our study among silage additives is related to the study by Chesson et al. (1985). According to this author, when there is variation

in this fraction between treatments, it is due to the natural selectivity of rumen microorganisms for different types of substrates. What did not happen with the inclusion level of 8% ground corn grain, therefore, is that this author's statement is consistent with our findings, because the use of corn did not increase the use of silage and consequently did not favor the microbial population of the rumen, not influencing the "I" fraction of NDF.

Filya (2003) evaluated the effect of strains of *Lactobacillus buchneri* and *Lactobacillus plantarum* alone or in combination on the ruminal degradability of corn and sorghum silages with low DM and stated that *L. buchneri*, *L. plantarum* or a combination of both did not influence the *in situ* ruminal degradability of DM, OM, and NDF of silages. These findings corroborate our results and those of the study conducted by Salawu et al. (2001), who observed that the application of *L. buchneri* to wheat and pea silage did not affect DM degradability.

However, the potential and effective degradability of NDF for silages containing IGCG was numerically higher when compared to NI silages and bacterial additives; however, it did not differ significantly between treatments, except for the potential degradability of silage treated with LBU1 compared to IGCG. If there is variation in the potential and effective degradability of NDF for silage with IGCG compared to NI and bacterial additives, this variation is due to the natural selectivity of rumen microorganisms for different types of substrates (Chesson et al. 1985). This author's statement is consistent with our findings, using corn increased the use of silage and consequently favored the microbial population in the rumen to increase NDF degradation and influencing silage degradation, although the degradation did not differ between treatments.

In summary, the effective and potential degradability of DM and OM was greater for silages containing IGCG compared to NI or bacterial additives. This finding can be explained by the greater degradability that maize presents in relation to elephant grass, because it has constituents that are easily soluble when compared to the BRS Capiaçu cultivar (Passini et al.,

2004). Passini et al. (2004) stated that the potential degradability of corn is 100%, regardless of how it is processed. This property explains the results found with the IGCG.

To classify a silage as high quality, knowledge of several factors is required when observing the fermentation profile (Kung et al., 2018). A high-quality silage must have a butyric acid content of less than 10 g/kg of DM, pH values between 3.5 and 4.6 for grasses, low DM loss, and ammoniacal N below 12% in relation to the total N (Borreani et al. 2018; Kung et al., 2018). Therefore, the silage containing IGCG achieved some requirements, showing that the inclusion of 8% IGCG in BRS Capiaçu elephant grass silage was essential for the preservation of the material during the fermentation process when the DM was low.

## CONCLUSIONS

The silage supplemented with ground corn grain showed the best fermentative profile and nutritional value in relation to silages with different doses of bacterial additives for low DM grass. However, the level of inclusion of ground corn grains was not efficient in reducing butyric fermentation at 60 and 90 d of fermentation, at the levels recommended by the literature. Regarding the *in situ* ruminal degradability of the silage with IGCG opened at 60 d of fermentation, there were increases in the degradability of DM and OM and improvements in their fractions when compared to the silages supplemented with bacterial additives. However, NDF degradation was not improved with the inclusion level of 8% of ground corn grain, with no improvement in its fractions. Thus, the addition of ground corn grain to elephant grass may be an alternative for greater rapid and abundant availability of nutrients (DM and OM) to the ruminal system.

## ACKNOWLEDGMENTS

This work was supported in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES; Brasilia, DF, Brazil), Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq; Brasilia, DF, Brazil) and Fundação de Amparo à Pesquisa do Estado de Minas Gerais (FAPEMIG; Belo Horizonte, MG, Brazil), which are Brazilian institutions that support studies.

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**Table 1.** Chemical composition of elephant grass (BRS Capiaçu) in natura, with application of doses of bacterial additives or inclusion of 8% ground corn grain.

| Variable | Additives | | | | | |
|---|---|---|---|---|---|---|
| | NI[1] | $LBU_{0.5}$[2] | $LBU_1$[3] | $LPP_1$[4] | $LPP_2$[5] | IGCG[6] |
| pH | 5.86 | 5.83 | 5.83 | 5.88 | 5.92 | 5.83 |
| Composition (g/kg DM) | | | | | | |
| Dry matter | 133.28 | 137.50 | 137.86 | 139.60 | 135.84 | 192.47 |
| Neutral detergent fiber | 704.52 | 689.36 | 678.92 | 682.91 | 680.96 | 524.93 |
| Acid detergent fiber | 452.83 | 480.43 | 438.80 | 467.86 | 472.36 | 317.12 |
| Crude protein | 65.01 | 68.47 | 67.56 | 66.90 | 67.30 | 71.44 |
| Ether extract | 15.45 | 13.48 | 16.63 | 15.00 | 15.72 | 18.98 |
| Mineral matter | 112.41 | 110.77 | 101.85 | 106.38 | 109.60 | 76.96 |
| Lignin | 95.15 | 74.51 | 74.41 | 78.86 | 89.81 | 66.04 |
| Hemicellulose | 251.69 | 208.93 | 240.12 | 215.06 | 208.60 | 207.81 |
| Soluble carbohydrates | 101.47 | 101.80 | 101.38 | 110.15 | 110.39 | 119.90 |
| Non-fibrous carbohydrates | 102.60 | 117.91 | 135.04 | 128.81 | 126.42 | 307.69 |
| Insoluble protein in acid detergent | 5.01 | 5.66 | 5.38 | 5.29 | 5.32 | 4.36 |
| Buffer capacity, e.mg NaOH/100 g DM | 15.01 | 14.55 | 14.51 | 14.72 | 14.33 | 11.43 |

[1]NI – No inoculant.

[2]$LBU_{0.5}$ - 0.5 g/ton of a commercial product composed of *Lactobacillus buchneri*, with minimum guarantee levels of $1.0 \times 10^{11}$ CFU/g.

[3]$LBU_1$ - 1 g/ton of a commercial product composed of *Lactobacillus buchneri*, with minimum guarantee levels of $1.0 \times 10^{11}$ CFU/g.

[4]$LPP_1$ - 1 g/ton of a commercial product composed of *Lactobacillus plantarum*, with minimum guarantee levels of $5.0 \times 10^{10}$ CFU/g and *Pediococcus acidilactici*, with minimum guarantee levels of $1.5 \times 10^{10}$ CFU/g.

[5]$LPP_2$ - 2 g/ton; of a commercial product composed of *Lactobacillus plantarum*, with minimum guarantee levels of $5.0 \times 10^{10}$ CFU/g and *Pediococcus acidilactici*, with minimum guarantee levels of $1.5 \times 10^{10}$ CFU/g.

[6]IGCG - inclusion of 8% of ground corn grain (corn meal).

**Table 2**. Proportion of ingredients and composition of the experimental diet.

| Ingredients | Level of inclusion g/kg DM |
|---|---|
| Corn silage | 558.70 |
| Ground Corn Grain | 158.44 |
| Soybean meal | 136.82 |
| Cotton whole | 93.26 |
| Sodium bicarbonate | 15.45 |
| Urea | 10.06 |
| [1]Nutronmilk | 10.30 |
| Magnesium oxide | 7.72 |
| Dicalcium phosphate | 5.08 |
| Limestone | 3.06 |
| Sulfur | 1.03 |
| Virginiamycin | 0.15 |
| Analysed composition | |
| Dry matter | 340.7 |
| Organic matter | 922.45 |
| Neutral detergent fiber | 256.22 |
| Crude protein | 166.71 |
| Ether extract | 43.13 |
| NFC | 456.5 |

Nutronmilk= 240 g/kg calcium, 40 g/kg phosphor, 25 g/kg magnesium, 20 g/kg sulfur, 75 g/kg sodium, 45 g/kg cobalt.

**Table 3.** Effect of levels of bacterial additives or inclusion of ground corn grain (IGCG) on different fermentation days on the chemical composition (g/kg of DM) of elephant grass cv. BRS Capiacu.

| Variable | Fermentation time (d) | | | SEM | Additives | | | | | | SEM | P- value | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 30 | 60 | 90 | | NI[1] | $LBU_{0.5}$[2] | $LBU_1$[3] | $LPP_1$[4] | $LPP_2$[5] | IGCG[6] | | IA[7] | d | IA × d |
| DM | $158^{A}$ | $145^{B}$ | $144^{B}$ | 1.28 | $136^{b}$ | $142^{b}$ | $138^{b}$ | $143^{b}$ | $140^{b}$ | $193^{a}$ | 1.81 | <0.001 | <0.001 | 0.21 |
| CP | $59.6^{A}$ | $48.3^{B}$ | $38.4^{C}$ | 1.80 | $42.6^{b}$ | $46.7^{b}$ | $43.6^{b}$ | $46.6^{b}$ | $48.0^{b}$ | $65.3^{a}$ | 2.55 | <0.001 | <0.001 | 0.78 |
| NDF | $644^{B}$ | $654^{B}$ | $673^{A}$ | 4.80 | $700^{a}$ | $680^{a}$ | $692^{a}$ | $696^{a}$ | $681^{a}$ | $494^{b}$ | 6.79 | <0.001 | <0.001 | 0.93 |
| ADF | $432^{C}$ | $442^{B}$ | $474^{A}$ | 2.89 | $480^{a}$ | $467^{a}$ | $477^{a}$ | $476^{a}$ | $468^{a}$ | $327^{b}$ | 4.09 | <0.001 | <0.001 | 0.33 |
| PIAD[8] | $3.78^{B}$ | $4.06^{AB}$ | $4.46^{A}$ | 0.124 | $4.32^{a}$ | $4.28^{a}$ | $4.62^{a}$ | $4.23^{a}$ | $4.29^{a}$ | $2.84^{b}$ | 0.175 | <0.001 | 0.001 | 0.24 |
| Lignin | $79.3^{B}$ | $87.1^{A}$ | $78.9^{B}$ | 1.56 | $86.7^{a}$ | $82.4^{a}$ | $90.1^{a}$ | $84.4^{a}$ | $87.0^{a}$ | $60.0^{b}$ | 2.20 | <0.001 | <0.001 | 0.56 |
| Hemicellulose | 213 | 212 | 199 | 4.37 | $220^{a}$ | $213^{a}$ | $215^{a}$ | $219^{a}$ | $212^{a}$ | $167^{b}$ | 6.18 | <0.001 | 0.056 | 0.95 |
| NFC | $171^{A}$ | $166^{A}$ | $154^{B}$ | 3.28 | $121^{b}$ | $137^{b}$ | $134^{b}$ | $126^{b}$ | $136^{b}$ | $329^{a}$ | 4.64 | <0.001 | 0.002 | 0.15 |
| Ash | $99.2^{B}$ | $109.8^{A}$ | $113.4^{A}$ | 1.21 | $115^{a}$ | $112^{a}$ | $112^{a}$ | $109^{a}$ | $114^{a}$ | $82^{b}$ | 1.71 | <0.001 | <0.001 | 0.06 |
| Ethereal extract | $25.7^{A}$ | $22.2^{B}$ | $20.9^{B}$ | 0.756 | $21.4^{bc}$ | $23.6^{b}$ | $18.9^{c}$ | $22.2^{bc}$ | $21.2^{bc}$ | $30.2^{a}$ | 1.07 | <0.001 | <0.001 | 0.81 |

Note: Capital letters compare the times of fermentation and lowercase letters comparing six silages by the Tukey test (P<0.05).

[1]NI - No inoculant.

[2]$LBU_{0.5}$ - 0.5 g/ton of a commercial product composed of *Lactobacillus buchneri*, with minimum guarantee levels of $1.0 \times 10^{11}$ CFU/g.

[3]$LBU_1$ - 1 g/ton of a commercial product composed of *Lactobacillus buchneri*, with minimum guarantee levels of $1.0 \times 10^{11}$ CFU/g.

[4]$LPP_1$ - 1 g/ton of a commercial product composed of *Lactobacillus plantarum*, with minimum guarantee levels of $5.0 \times 10^{10}$ CFU/g and *Pediococcus acidilactici*, with minimum guarantee levels of $1.5 \times 10^{10}$ CFU/g.

[5]$LPP_2$ - 2 g/ton; of a commercial product composed of *Lactobacillus plantarum*, with minimum guarantee levels of $5.0 \times 10^{10}$ CFU/g and *Pediococcus acidilactici*, with minimum guarantee levels of $1.5 \times 10^{10}$ CFU/g.

[6]IGCG - inclusion of 8% of ground corn grain (cornmeal).

[7] Additives.

[8]PIAD- Acid detergent insoluble protein.

**Table 4**. Effect of levels of bacterial additives or inclusion of ground corn grain and different fermentation days on pH values, soluble carbohydrates (g/kg DM), lactic acid/acetic acid ratio (g/kg DM), production of effluents (kg/ton of green mass) and total DM losses (% of DM).

| Variable | Fermentation time (d) | | | SEM | Additives | | | | | | SEM | P-value | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 30 | 60 | 90 | | NI[1] | $LBU_{0.5}$[2] | $LBU_1$[3] | $LPP_1$[4] | $LPP_2$[5] | IGCG[6] | | IA[7] | d | IA× d |
| pH | $4.06^{B}$ | $4.92^{A}$ | $4.87^{A}$ | 0.095 | $4.81^{a}$ | $4.64^{a}$ | $4.78^{a}$ | $4.80^{a}$ | $4.66^{a}$ | $3.99^{b}$ | 0.135 | <0.001 | <0.001 | 0.14 |
| WSC[8] | $4.81^{B}$ | $5.57^{A}$ | $5.52^{A}$ | 0.141 | 5.53 | 5.20 | 5.29 | 4.99 | 5.22 | 5.58 | 0.199 | 0.32 | <0.001 | 0.20 |
| L/A ratio[9] | $2.64^{A}$ | $0.51^{B}$ | $0.51^{B}$ | 0.192 | $0.832^{b}$ | $0.773^{b}$ | $0.550^{b}$ | $1.046^{b}$ | $0.736^{b}$ | $3.375^{a}$ | 0.158 | <0.001 | <0.001 | 0.52 |
| EP[10] | 89.4 | 88.4 | 87.6 | 2.18 | $85.3^{b}$ | $88.1^{ab}$ | $85.5^{ab}$ | $89.2^{ab}$ | $98.3^{a}$ | $85.0^{b}$ | 3.09 | 0.03 | 0.83 | 0.81 |
| Total DM losses | $3.09^{B}$ | $10.81^{A}$ | $11.58^{A}$ | 0.655 | 8.41 | 6.78 | 9.84 | 7.87 | 8.90 | 9.17 | 0.927 | 0.26 | <0.001 | 0.10 |

Note: Capital letters compare the times of fermentation and lowercase letters comparing six silages by the Tukey test (P<0.05).

[1]NI- No inoculant.

[2]$LBU_{0.5}$ - 0.5 g/ton of a commercial product composed of *Lactobacillus buchneri*, with minimum guarantee levels of $1.0 \times 10^{11}$ CFU/g.

[3]$LBU_1$ - 1 g/ton; of a commercial product composed of *Lactobacillus buchneri*, with minimum guarantee levels of $1.0 \times 10^{11}$ CFU/g.

[4]$LPP_1$ - 1 g/ton of a commercial product composed of *Lactobacillus plantarum*, with minimum guarantee levels of $5.0 \times 10^{10}$ CFU/g and *Pediococcus acidilactici*, with minimum guarantee levels of $1.5 \times 10^{10}$ CFU/g.

[5]$LPP_2$ - 2 g/ton; of a commercial product composed of *Lactobacillus plantarum*, with minimum guarantee levels of $5.0 \times 10^{10}$ CFU/g and *Pediococcus acidilactici*, with minimum guarantee levels of $1.5 \times 10^{10}$ CFU/g.

[6]IGCG - inclusion of 8% of ground corn grain (cornmeal).

[7]IA- Additives.

[8]WSC- Water soluble carbohydrates.

[9]L/A ratio- Lactic and acetic acid ratio.

[10]EP- Effluent production.

**Table 5.** Effect of levels of bacterial additives or inclusion of ground corn grain (IGCG) on different fermentation days on ammoniacal nitrogen, gas loss and organic acid of elephant grass cv. BRS Capiacu.

| Variable | Fermentation time (d) | | | | | | SEM | P-value | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | 30 | | | | | | | | | |
| | NI[1] | $LBU_{0.5}$[2] | $LBU_1$[3] | $LPP_1$[4] | $LPP_2$[5] | IGCG[6] | | IA[7] | d | IA x d |
| Ammoniacal nitrogen, % TN | $16.99^{aB}$ | $13.41^{aC}$ | $17.31^{aC}$ | $14.73^{aB}$ | $13.55^{aB}$ | $8.56^{aA}$ | 3.91 | <0.001 | <0.001 | <0.001 |
| Gas loss, % of DM | $4.69^{aC}$ | $4.40^{aB}$ | $5.65^{aB}$ | $4.59^{aB}$ | $4.57^{aB}$ | $3.58^{aA}$ | 1.10 | <0.001 | <0.001 | 0.001 |
| Lactic acid, g/kg DM | $14.92^{bcA}$ | $16.74^{abA}$ | $12.97^{cA}$ | $17.51^{abA}$ | $15.65^{bcA}$ | $19.51^{aA}$ | 0.885 | <0.001 | <0.001 | <0.001 |
| Acetic acid, g/kg DM | $6.19^{abB}$ | $7.52^{abC}$ | $8.48^{aB}$ | $6.40^{abC}$ | $7.71^{abC}$ | $4.36^{bA}$ | 0.825 | <0.001 | <0.001 | <0.001 |
| Propionic acid, g/kg DM | $1.03^{aC}$ | $0.893^{aC}$ | $1.18^{aB}$ | $0.548^{aB}$ | $1.04^{aB}$ | $0.602^{aA}$ | 0.166 | <0.001 | <0.001 | <0.001 |
| Butiric acid, g/kg DM | $26.90^{aB}$ | $15.00^{bcC}$ | $29.90^{aB}$ | $18.10^{bB}$ | $11.20^{cB}$ | $12.10^{cC}$ | 1.17 | <0.001 | <0.001 | <0.001 |
| | 60 | | | | | | | | | |
| Ammoniacal nitrogen, % TN | $54.85^{aA}$ | $27.97^{bcB}$ | $40.96^{abB}$ | $38.49^{abA}$ | $38.36^{abA}$ | $19.41^{cA}$ | 3.91 | <0.001 | <0.001 | <0.001 |
| Gas loss, % of DM | $10.26^{aB}$ | $9.96^{aA}$ | $10.25^{aA}$ | $10.04^{aA}$ | $7.46^{aAB}$ | $5.74^{aA}$ | 1.10 | <0.001 | <0.001 | 0.001 |
| Lactic acid, g/kg DM | $0.600^{bB}$ | $0.945^{bB}$ | $0.492^{bB}$ | $0.562^{bB}$ | $3.18^{bB}$ | $14.59^{aB}$ | 0.885 | <0.001 | <0.001 | <0.001 |
| Acetic acid, g/kg DM | $17.03^{bcA}$ | $19.89^{bA}$ | $15.64^{cA}$ | $19.36^{bA}$ | $25.57^{aA}$ | $5.26^{dA}$ | 0.825 | <0.001 | <0.001 | <0.001 |
| Propionic acid, g/kg DM | $2.20^{abB}$ | $1.72^{bB}$ | $2.78^{aA}$ | $2.07^{bA}$ | $1.84^{bA}$ | $0.958^{cA}$ | 0.166 | <0.001 | <0.001 | <0.001 |
| Butiric acid, g/kg DM | $33.70^{bA}$ | $22.50^{deB}$ | $42.10^{aA}$ | $19.10^{eB}$ | $27.80^{cA}$ | $26.30^{cdA}$ | 1.17 | <0.001 | <0.001 | <0.001 |
| | 90 | | | | | | | | | |
| Ammoniacal nitrogen, % TN | $65.35^{aA}$ | $57.06^{abA}$ | $58.91^{abA}$ | $48.10^{bA}$ | $45.04^{bA}$ | $13.71^{cA}$ | 3.91 | <0.001 | <0.001 | <0.001 |
| Gas loss, % of DM | $17.97^{aA}$ | $9.92^{bA}$ | $9.85^{bA}$ | $8.29^{bAB}$ | $8.44^{bA}$ | $6.30^{bA}$ | 1.10 | <0.001 | <0.001 | 0.001 |
| Lactic acid, g/kg DM | $0.538^{bB}$ | $0.507^{bB}$ | $0.609^{bB}$ | $2.69^{bB}$ | $0.757^{bB}$ | $12.69^{aB}$ | 0.885 | <0.001 | <0.001 | <0.001 |
| Acetic acid, g/kg DM | $17.48^{aA}$ | $13.77^{bB}$ | $13.54^{bA}$ | $15.25^{abB}$ | $15.27^{abB}$ | $4.70^{cA}$ | 0.825 | <0.001 | <0.001 | <0.001 |
| Propionic acid, g/kg DM | $5.12^{aA}$ | $5.09^{aA}$ | $3.02^{bA}$ | $2.11^{cA}$ | $2.11^{cA}$ | $0.965^{dA}$ | 0.166 | <0.001 | <0.001 | <0.001 |
| Butiric acid, g/kg DM | $33.50^{aA}$ | $27.20^{bA}$ | $29.60^{abB}$ | $34.00^{aA}$ | $29.50^{abA}$ | $21.20^{cB}$ | 1.17 | <0.001 | <0.001 | <0.001 |

Note: Lower case letters comparing additives at 30, 60, and 90 d and capital letters comparing 30, 60, and 90 d for each additive by the Tukey test ($P<0.05$).

[1]NI- No inoculant.

[2]$LBU_{0.5}$ - 0.5 g/ton of a commercial product composed of *Lactobacillus buchneri*, with minimum guarantee levels of $1.0 \times 10^{11}$ CFU/g.

[3]$LBU_1$ - 1 g/ton; of a commercial product composed of *Lactobacillus buchneri*, with minimum guarantee levels of $1.0 \times 10^{11}$ CFU/g.

[4]$LPP_1$ - 1 g/ton of a commercial product composed of *Lactobacillus plantarum*, with minimum guarantee levels of $5.0 \times 10^{10}$ CFU/g and *Pediococcus acidilactici*, with minimum guarantee levels of $1.5 \times 10^{10}$ CFU/g.

[5]$LPP_2$ - 2 g/ton; of a commercial product composed of *Lactobacillus plantarum*, with minimum guarantee levels of $5.0 \times 10^{10}$ CFU/g and *Pediococcus acidilactici*, with minimum guarantee levels of $1.5 \times 10^{10}$ CFU/g.

[6]IGCG - inclusion of 8% of ground corn grain (cornmeal).

[7]IA- Additives.

**Table 6.** Effect of levels of bacterial additives or inclusion of ground corn grain (IGCG) on *in situ* degradation parameters of DM, OM and NDF of elephant grass silages cv. BRS Capiaçu.

| Items | Additives | | | | | | SEM | P-value |
|---|---|---|---|---|---|---|---|---|
| | NI[1] | $LBU_{0.5}$[2] | $LBU_1$[3] | $LPP_1$[4] | $LPP_2$[5] | IGCG[6] | | IA[7] |
| DM parameters | | | | | | | | |
| a[8], g/kg | 224[c] | 248[b] | 222[c] | 243[bc] | 232[bc] | 419[a] | 4.87 | <0.001 |
| b[9], g/kg | 451[a] | 436[a] | 451[a] | 440[a] | 457[a] | 361[b] | 6.37 | <0.001 |
| c[10], %/per $h^{-1}$ | 0.0191 | 0.0197 | 0.0208 | 0.0182 | 0.0194 | 0.0244 | 0.00191 | 0.32 |
| OM parameters | | | | | | | | |
| a[8], g/kg | 179[bc] | 197[b] | 166[c] | 188[bc] | 175[bc] | 400[a] | 5.42 | <0.001 |
| b[9], g/kg | 474[a] | 464[a] | 481[a] | 470[a] | 491[a] | 370[b] | 7.51 | <0.001 |
| c[10], %/per $h^{-1}$ | 0.0193 | 0.0199 | 0.0211 | 0.0186 | 0.0195 | 0.0246 | 0.002 | 0.37 |
| NDF parameters | | | | | | | | |
| D[11], g/kg | 540[ab] | 528[b] | 530[b] | 535[ab] | 559[ab] | 578[a] | 10.2 | 0.029 |
| C[12], %/per $h^{-1}$ | 0.0183 | 0.0189 | 0.0196 | 0.0182 | 0.0182 | 0.0217 | 0.00197 | 0.79 |
| I[13], g/kg | 385 | 384 | 387 | 377 | 369 | 357 | 6.56 | 0.050 |

Note: Means with superscript letters on the lines differ additives by the Tukey test (P<0.05).

[1]NI- No inoculant.

[2]$LBU_{0.5}$ - 0.5 g/ton of a commercial product composed of *Lactobacillus buchneri*, with minimum guarantee levels of $1.0 \times 10^{11}$ CFU/g.

[3]$LBU_1$ - 1 g/ton; of a commercial product composed of *Lactobacillus buchneri*, with minimum guarantee levels of $1.0 \times 10^{11}$ CFU/g.

[4]$LPP_1$ - 1 g/ton of a commercial product composed of *Lactobacillus plantarum*, with minimum guarantee levels of $5.0 \times 10^{10}$ CFU/g and *Pediococcus acidilactici*, with minimum guarantee levels of $1.5 \times 10^{10}$ CFU/g.

[5]$LPP_2$ - 2 g/ton; of a commercial product composed of *Lactobacillus plantarum*, with minimum guarantee levels of $5.0 \times 10^{10}$ CFU/g and *Pediococcus acidilactici*, with minimum guarantee levels of $1.5 \times 10^{10}$ CFU/g.

[6]IGCG - inclusion of 8% of ground corn grain (cornmeal).

[7]IA- Additives.

[8]a- Is the water soluble fraction of the feed at time zero (g/kg DM).

[9]b- Potentially degradable fraction in the rumen (g/kg DM)

[10]c- Is the degradation rate of the potentially degradable fraction in the rumen (b) (%/per $h^{-1}$).

[11]D- NDF fraction potentially degradable in the rumen (g/kg DM).

[12]C- Degradation rate of the fraction D (%/per $h^{-1}$).

[13]I- Non-degradable fraction of the NDF (g/kg DM).

**Table 7.** Effect of levels of bacterial additives or inclusion of ground corn grain (IGCG) on potential degradability of DM, OM and NDF of elephant grass silages cv. BRS Capiaçu.

| Items | Additives | | | | | | SEM | P-value |
|---|---|---|---|---|---|---|---|---|
| | NI[1] | $LBU_{0.5}$[2] | $LBU_1$[3] | $LPP_1$[4] | $LPP_2$[5] | IGCG[6] | | IA[7] |
| Potential degradability of DM, g/kg | 675[b] | 684[b] | 673[b] | 682[b] | 689[b] | 780[a] | 4.77 | <0.001 |
| Potential degradability of OM, g/kg | 653[b] | 661[b] | 647[b] | 658[b] | 666[b] | 770[a] | 4.79 | <0.001 |
| Potential degradability of NDF, g/kg | 584[ab] | 579[ab] | 578[b] | 587[ab] | 603[ab] | 618[a] | 8.45 | 0.034 |

Note: Means with superscript letters on the lines differ additives by the Tukey test (P<0.05).

[1]NI- No inoculant.

[2]$LBU_{0.5}$ - 0.5 g/ton of a commercial product composed of *Lactobacillus buchneri*, with minimum guarantee levels of $1.0 \times 10^{11}$ CFU/g.

[3]$LBU_1$ - 1 g/ton; of a commercial product composed of *Lactobacillus buchneri*, with minimum guarantee levels of $1.0 \times 10^{11}$ CFU/g.

[4]$LPP_1$ - 1 g/ton of a commercial product composed of *Lactobacillus plantarum*, with minimum guarantee levels of $5.0 \times 10^{10}$ CFU/g and *Pediococcus acidilactici*, with minimum guarantee levels of $1.5 \times 10^{10}$ CFU/g.

[5]$LPP_2$ - 2 g/ton; of a commercial product composed of *Lactobacillus plantarum*, with minimum guarantee levels of $5.0 \times 10^{10}$ CFU/g and *Pediococcus acidilactici*, with minimum guarantee levels of $1.5 \times 10^{10}$ CFU/g.

[6]IGCG - inclusion of 8% of ground corn grain (cornmeal).

[7]IA- Additives.

**Table 8.** Effect of levels of bacterial additives or inclusion of ground corn grain (IGCG) on effective degradability of DM, OM and NDF of elephant grass silages cv. BRS Capiaçu.

| Items | Additives | | | | | | SEM | P-value |
|---|---|---|---|---|---|---|---|---|
| | NI[1] | $LBU_{0.5}$[2] | $LBU_1$[3] | $LPP_1$[4] | $LPP_2$[5] | IGCG[6] | | IA[7] |
| Effective DM degradability, g/kg | | | | | | | | |
| Passage fee (%/$h^{-1}$) | | | | | | | | |
| 2 | 443[b] | 463[b] | 450[b] | 451[b] | 457[b] | 616[a] | 10 | <0.001 |
| 5 | 348[b] | 371[b] | 354[b] | 360[b] | 360[b] | 537[a] | 8.73 | <0.001 |
| 8 | 310[b] | 334[b] | 315[b] | 324[b] | 321[b] | 503[a] | 7.41 | <0.001 |
| Effective OM degradability, g/kg | | | | | | | | |
| Passage fee (%/$h^{-1}$) | | | | | | | | |
| 2 | 411[b] | 427[b] | 411[b] | 413[b] | 418[b] | 603[a] | 10.60 | <0.001 |
| 5 | 311[b] | 328[b] | 308[b] | 315[b] | 313[b] | 522[a] | 9.33 | <0.001 |
| 8 | 271[b] | 289[b] | 266[b] | 277[b] | 272[b] | 487[a] | 7.93 | <0.001 |
| Effective NDF degradability, g/kg | | | | | | | | |
| Passage fee (%/$h^{-1}$) | | | | | | | | |
| 2 | 278 | 279 | 284 | 277 | 287 | 320 | 13.40 | 0.26 |
| 5 | 156 | 157 | 162 | 155 | 161 | 186 | 11.10 | 0.40 |
| 8 | 108 | 110 | 113 | 108 | 112 | 131 | 8.72 | 0.44 |

Note: Means with superscript letters on the lines differ additives by the Tukey test (P<0.05).

[1]NI- No inoculant.

[2]$LBU_{0.5}$ - 0.5 g/ton of a commercial product composed of *Lactobacillus buchneri*, with minimum guarantee levels of $1.0 \times 10^{11}$ CFU/g.

[3]$LBU_1$ - 1 g/ton; of a commercial product composed of *Lactobacillus buchneri*, with minimum guarantee levels of $1.0 \times 10^{11}$ CFU/g.

[4]$LPP_1$ - 1 g/ton of a commercial product composed of *Lactobacillus plantarum*, with minimum guarantee levels of $5.0 \times 10^{10}$ CFU/g and *Pediococcus acidilactici*, with minimum guarantee levels of $1.5 \times 10^{10}$ CFU/g.

[5]$LPP_2$ - 2 g/ton; of a commercial product composed of *Lactobacillus plantarum*, with minimum guarantee levels of $5.0 \times 10^{10}$ CFU/g and *Pediococcus acidilactici*, with minimum guarantee levels of $1.5 \times 10^{10}$ CFU/g.

[6]IGCG - inclusion of 8% of ground corn grain (cornmeal).

[7]IA- Additives.

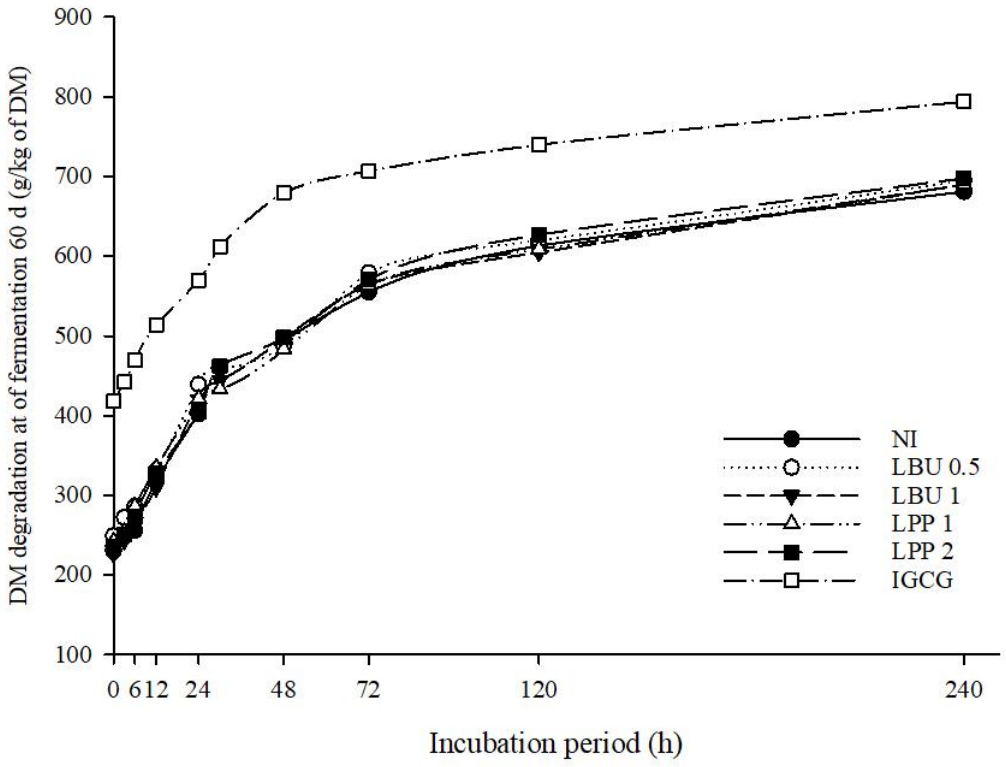


**Figure 1**. Total degradability of dry matter of elephant grass silages with doses of bacterial additives or inclusion of ground corn grains. Estimated by the equation:

$Deg(t) = a + b\ (1 - e^{(-ct)})$.

* Deg (t)= represents the degradability or disappearance of the constituent (DM) of the feed; a = is the water-soluble fraction of the feed at time zero; b = is the fraction insoluble in water, but potentially degradable in the rumen at a given time; c = is the degradation rate of the potentially degradable fraction in the rumen (b); t = is the incubation time (h).

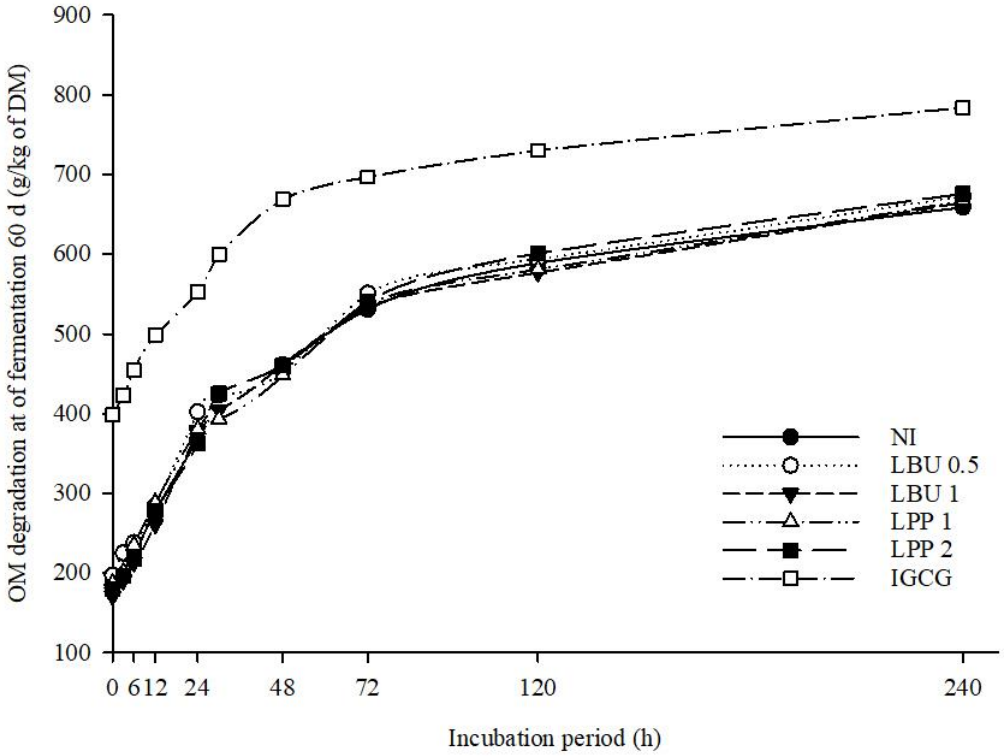


**Figure 2**. Total degradability of organic matter in elephant grass silages with doses of bacterial additives or ground corn grain. Estimated by the equation:

$Deg(t) = a + b\ (1 - e^{(-ct)})$.

* Deg (t)= represents the degradability or disappearance of the constituent (OM) of the feed; a = is the water-soluble fraction of the feed at time zero; b = is the fraction insoluble in water, but potentially degradable in the rumen at a given time; c = is the degradation rate of the potentially degradable fraction in the rumen (b); t = is the incubation time (h).

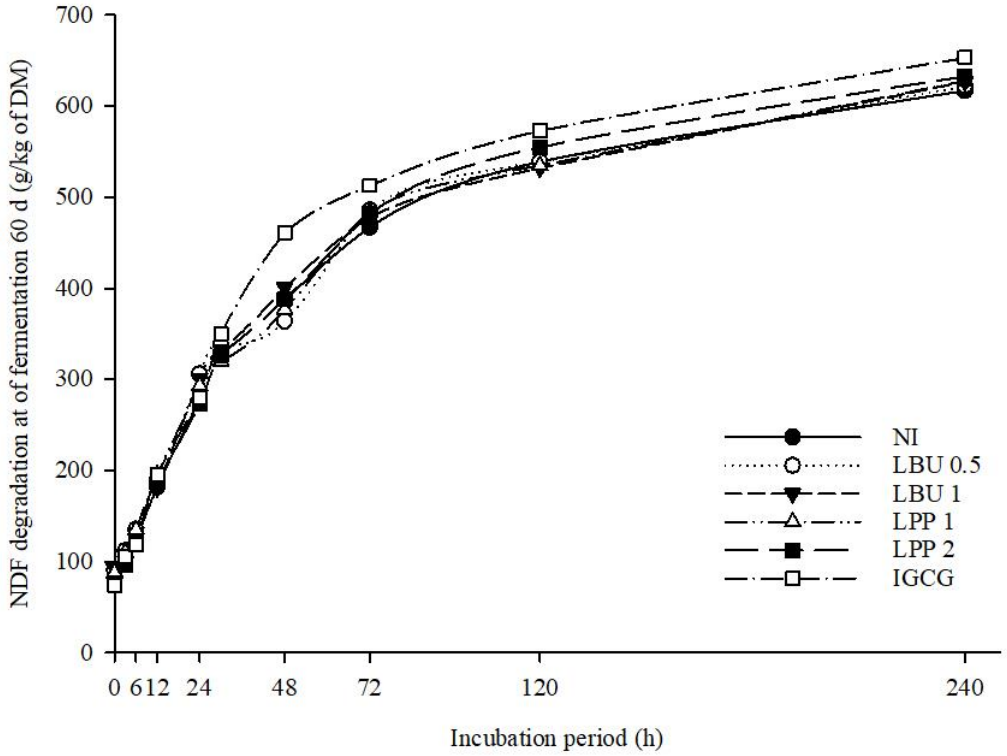


**Figure 3**. Total degradability of neutral detergent fiber of elephant grass silages with doses of bacterial additives or ground corn grain. Estimated by the equation:

$$R(t) = D\ (e^{-ct}) + I.$$

*R(t)= is the incubation residue at time t (h); D= is the fraction of potentially degradable NDF in the rumen; c= is the rate of degradation of fraction D; I= is the non-degradable fraction of NDF.