

Losses, chemical composition and aerobic stability of rye silage cv. Temprano subjected to different pre-flowering cuttings with or without fungicide

Perdas, composição química e estabilidade aeróbia da silagem de centeio cv. Temprano submetido a diferentes sistemas de corte com ou sem fungicida no pré-florescimento

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Highlights:

Silages of rye harvested at the vegetative stage presented better fiber composition, lower lignification.

Silages of rye harvested at the vegetative stage presented a less efficient fermentation process, lower concentration of organic acids and aerobic stability.

The use of fungicide resulted in better dry matter content and fiber composition and improved the fermentation process generating lower DM losses.

Abstract

The present study evaluated the fermentation, chemical characteristics and ruminal disappearance of dry matter of silages of rye cv. Temprano harvested at the stage of floury grain, managed with or without cutting at the vegetative stage and with or without application of Fluxapyroxad + Pyraclostrobin-based fungicide at the pre-flowering phenological stage. This was a 2x2 factorial randomized block experimental design, with 2 cutting regimes (0 and 1 cut) and 2 application management (with and without fungicide), with six replications, where each repetition is represented by a plot of 9.45m², with the treatments SC-SF: without cut at the vegetative stage and without fungicide application; SC-CF: without cut at the vegetative stage with fungicide application; UC-SF: a cut at the vegetative stage with pre-flowering fungicide application; UC-CF: a cut at the vegetative stage and without fungicide application. UC-CF silages were better in the general context, due to the better fiber composition with an average 6.75% reduction in neutral detergent fiber and acid detergent fiber and a 17.10% reduction in lignin content, but, due to its higher DM content, it presented a less efficient fermentation process in terms of organic acid concentration and aerobic stability. However, it did not change the DM losses or ruminal degradability, and there is also the use of DM harvested at the vegetative stage that certainly has a superior quality. The use of fungicide provided only better dry matter index and better fiber composition and improved the fermentation process resulting in lower DM losses without altering the aerobic stability of silages.

Key words: Organic acids. Ruminal disappearance. Potential of hydrogen. *Secale cereale*.

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Resumo

Objetivou-se avaliar as características fermentativas, bromatológicas e o desaparecimento ruminal da matéria seca das silagens de Centeio cv. Temprano colhidas no estágio de grão farináceo, manejado com ou sem corte no estágio vegetativo e com ou sem aplicação de fungicida a base de Fluxapiraxade + Piraclostrobina no estágio fenológico de pré-florescimento. O delineamento experimental foi o de blocos ao acaso em esquema fatorial 2x2, sendo 2 regimes de cortes (0 e 1 corte) e 2 manejos de aplicação (com e sem fungicida), com seis repetições, onde cada repetição é representada por uma parcela de 9,45m², sendo os tratamentos SC-SF: sem corte no vegetativo e sem aplicação de fungicida; SC-CF: sem corte no vegetativo com aplicação de fungicida; UC-SF: um corte no vegetativo com aplicação de fungicida no pré-florescimento; UC-CF: um corte no vegetativo e sem aplicação de fungicida; As silagens do tratamento UC-CF apresentaram-se melhores no contexto geral, devido a melhor composição da fibra com redução de 6,75% em média nos teores de fibra em detergente neutro e fibra em detergente ácido e uma redução de 17,10% nos teores de lignina, no entanto devido ao seu maior teor de MS apresentou um processo fermentativo menos eficiente em termos de concentração de ácidos orgânicos e de estabilidade aeróbia, porém, o mesmo não alterou as perdas de MS nem degradabilidade ruminal, atribuído a isso ainda tem-se o aproveitamento da MS colhida no estágio vegetativo que tem certamente uma qualidade superior. A utilização de fungicida proporcionou apenas melhores índices de matéria seca e melhor composição da fibra e, melhorou o processo fermentativo gerando menores perdas de MS sem alterar a estabilidade aeróbica das silagens.

Palavras-chave: Ácidos orgânicos. Desaparecimento ruminal. Potencial hidrogeniônico. *Secale cereale*.

Introduction

The southern Brazil has stood out in the national scenario due to the efficiency of integrated agricultural production systems, where rotations are performed between annual grain crops in summer and, in winter, with plants and/or pastures previously intended for only soil cover, however in current scenarios, they are increasingly being used for animal feed (Fontanelli et al., 2016).

According to Fontaneli, Santos, Fontaneli, Hentz and Lehmen (2011), forages important for cultivation include Black Oats and Ryegrass, seconded by cereals that can be used in dual purpose systems such as wheat, White Oats, Barley, Triticale and Rye. These materials have been shown to be very versatile and can be used in animal nutrition at various stages of development, at the vegetative stage used for direct foraging (Nascimento et al., 2013). Also, according to Bumbieris et al. (2011), its surplus can be used for the production of hay and haylage and to have food stocked for other times, and the crop can be used more often, since

for haylage production, it is harvested before completing its normal cycle. And, finally, the use at the reproductive stages for grain production and its participation in concentrates or for production of silage of the whole plant (Bortolini, Sandini, Carvalho, & Moraes, 2004).

Ensiling, according to Pereira and Reis (2001), is a process with the objective of preserving fresh forage, from anaerobic fermentation, which occurs basically due to the conversion of soluble carbohydrates from plants or added into organic acids, due to the action of various microorganisms that, in the proper environment, proliferate and provide adequate conditions for the preservation of the original nutritional value.

According to Baier (1994), rye is an annual winter plant belonging to the family of grasses, which has a fasciculate and aggressive root system, has a clumping growth habit, has erect, glabrous, cylindrical stems with structure varying from 1.2 to 1.8 m in height, its linear bluish-green leaves with membranous ligules and small auricles.

Winter cereal silage becomes an alternative to supply quality roughage in periods where there is low supply in the field, as well as allowing the partial replacement of traditional silages, such as corn and sorghum, which have a higher production cost and not competing with traditional summer crops, such as soybeans (Zamarchi, Pavinato, Menezes, & Martin, 2014)

One of the obstacles to the production of winter cereals in southern Brazil is the climatic conditions usually encountered during the crop cycle, mainly due to excessive rainfall, which is a determinant factor in favoring the increase of diseases (Reis & Casa, 2007). For Goulart (1998), a tool that has the ability to stabilize productivity is the correct use of different classes of fungicides.

In order to properly adjust the supply of feed that makes up the diet of domestic animals, the digestive behavior evaluations have had a considerable development in recent years; the best results of animal productivity are directly associated with the quality of the feed provided. Forage has a major role in livestock production systems, providing fiber, which is generally a low energy source, and is responsible for maintaining rumen function, in addition to regulating intake and supplying important compounds for the production of milk and meat (Carvalho, Santos, Gonçalves, Moraes, & Nabinger, 2010).

From this assumption, the goal of this study was to evaluate the fermentation, chemical characteristics and ruminal disappearance of dry matter of silages of rye cv. Temprano, managed with or without cutting at the vegetative stage and with or without application of Fluxapyroxad + Pyraclostrobin-based fungicide at the pre-flowering phenological stage.

Material and Methods

The experiment was conducted at the Animal Production Center ((Núcleo de Produção Animal – NUPRAN) belonging to the Agrarian and

Environmental Sciences Sector of the Universidade Estadual do Centro-Oeste (UNICENTRO), located in Guarapuava, State of Paraná, located in the subtropical zone of the State of Paraná (Maack, 2002), at the geographic coordinates 25°23'02" South latitude and 51°29'43" West longitude and 1,026 m altitude.

The climate of the region according to the Köppen classification is Cfb (mesothermal humid subtropical), with mild summers and moderate winter, no well-defined dry season and with severe frosts. It is characterized by an average annual rainfall of 2,022 mm and an average annual temperature of 16.5°C (Nitsche, Caramori, Ricce, & Pinto, 2019).

The soil of the experimental area was classified as Typic Latosol Bruno (Pott, Müller, & Bertelli, 2007). Before planting, the area had the following chemical characteristics (0 to 20 cm profile): pH 0.01M CaCl₂: 4.8; phosphorus: 1.0 mg dm⁻³; K⁺: 0.2 cmol_c dm⁻³; OM: 2.62%; Al³⁺: 0.0 cmol_c dm⁻³; H⁺+Al³⁺: 5.2 cmol_c dm⁻³; Ca²⁺: 5.2 cmol_c dm⁻³; Mg²⁺: 5.1 cmol_c dm⁻³ and base saturation: 65.8%.

The experimental material used was Rye (*Secale cereale*), cultivar Temprano. The treatments consisted of silages resulting from the harvest at the stage of flouy grain, being SC-SF: without cut and without fungicide application; SC-CF: without cut with fungicide application; UC-SF: a cut at the vegetative stage with pre-flowering fungicide application; UC-CF: a cut at the vegetative stage and without fungicide application.

The experimental field consisted of a total area of 540 m², distributed in 24 plots of 9.45 m² each (3.15m x 3m) corresponding to 54 linear meters of sowing, where the useful area of 4.2 m² (2.1m x 2m) corresponded to 24 linear meters of sowing. The experiment was sown under no-till system on April 12, 2018 evenly for the treatments. At planting, the row spacing was 0.175 m, the sowing depth was 0.02 m with an average distribution of 220 seeds per m².

Upon rye implantation, basal fertilization was performed with 280 kg ha⁻¹ fertilizer 08-30-20 (N-P₂O₅-K₂O), following the recommendations of the manual of fertilization and liming for the State of Paraná (Sociedade Brasileira de Ciência do Solo, Núcleo Estadual do Paraná [SBCS/NEPAR], 2017). Nitrogen fertilization was performed 51 days after sowing and consisted of a single application of 444 kg ha⁻¹ urea (45-00-00), ensuring the application of 200 kg ha⁻¹ nitrogen.

Chemical weed control with herbicides was primarily carried out in the desiccation of the experimental area before sowing, using glyphosate-based herbicide (Roundup Original[®] commercial product: 2.0 L ha⁻¹); during crop management, 30 days after sowing, metsulfuron-methyl herbicide was applied (Ally[®] commercial product: 6.6 g ha⁻¹). Pest control was carried out 30 days after sowing, according to field evaluations, using Thiamethoxam + Lambda-cyhalothrin insecticide (Engeo Pleno[®] commercial product: 150 mL ha⁻¹) and another application 145 days after sowing, according to field evaluation.

The Fluxapyroxad + Pyraclostrobin-based fungicide (OrkestraTMSC[®] commercial product: 0.35 L ha⁻¹) was applied at pre-flowering in treatments with fungicide application.

The management regime of treatments subjected to a cut (T3 and T4) was performed in the stages of full tillering. The lowering height followed the recommendations of Fontaneli, Santos, Nascimento Jr, Minella and Caierão (2009), that is, 0.10m from the soil level.

Silages were made in equivalence of reproductive stages, being defined the stage of floury grain, the indicated time for ensiling, according to Fontaneli et al. (2009), being the cuts made on 188 days, 195 days after according to treatment of 0 or 1 cut, respectively.

The cutting of the plants in the useful area of each plot (4.2 m²) was done manually with the aid of a serrated sickle. Upon ensiling, the plants

of each plot were processed in a stationary forage chopper, model EM 6400, Nogueira[®], and ensiled in experimental silos. After processing, the material should have an average particle size of 3.7cm, determined according to the methodology proposed by Jobim, Nussio, Reis and Schmidt (2007). The silos were PVC pipes, 10cm in diameter, 50cm in height (0.003m³ volume) and capacity for about 3kg, manually compacted and properly labeled. Silos were sealed with black tarpaulin and adhesive tape.

Silos were opened, on average, 60 days after sealing, two representative samples of silages were collected from each silo, the first for aerobic stability evaluations and the second for analysis of dry matter contents, drying the material in a forced air oven at 55°C to constant weight between weighings (Association of Official Analytical Chemists [AOAC], 1984).

Aerobic stability was evaluated by temperature and pH measurements. From each experimental silo, a 400g sample was taken and placed in 1kg buckets. These were stored in a controlled air-conditioned environment, with a temperature programmed to remain stable at 25°C throughout the evaluation time. To determine aerobic stability, temperature and pH were read daily (at 7h, 15h and 23h), and the evaluation time was maintained until the aerobic stability of the material was lost.

Silage temperature was measured using a Gulterm 1001 long stem digital thermometer inserted at the center of the forage mass, pH readings were made using a digital potentiometer, according to the methodology established by Cherney and Cherney (2003).

The criterion for defining aerobic stability break was considered when the pH increased at levels above 0.5 units, as mentioned by Weinberg et al. (2007).

All pre-dried silage samples were ground to 1mm in a Wiley mill, where sequentially determinations were performed for total dry matter in an oven at

105°C for 16h, crude protein (CP) by micro Kjeldahl method, mineral matter (MM) by incineration at 550°C (4h) (Silva & Queiroz, 2009). Contents of neutral detergent fiber (NDF) was determined according to Van Soest, Robertson and Lewis (1991) using thermostable α amylase (Termamyl 120L, Novozymes Latin America Ltda.), acid detergent fiber (ADF) and lignin (LIG), according to Goering and Van Soest (1970) and the contents of Hemicellulose (HEM) and Cellulose (CEL) by the equation (HEM = NDF - ADF and CEL = ADF - LIG) following the methodology proposed by Silva and Queiroz (2009).

Ruminal disappearance of DM from silages was estimated by the in situ degradability technique using 12x8cm nylon bags with 40 to 60 μ m pores, containing approximately 5g each material, ground to 1mm, for subsequent rumen incubation (Nocek, 1988). The incubation times used were 24 and 48 hours. This procedure was previously subjected for consideration and approval by the Animal Use Ethics Committee of UNICENTRO, under opinion 035/2017 CEUA/UNICENTRO. For this, we used two 48-month-old Jersey steers housed in the beef cattle didactic unit, with an average body weight of 550 kg and with ruminal fistula implanted by the rumenotomy technique, previously approved by the Animal Use Ethics Committee of UNICENTRO, under opinion 030/2014 - CEUA/UNICENTRO.

The lactic acid concentration was determined according to the methodology described by Pryce (1969). Ethanol, acetic, propionic and butyric acid concentrations in the samples were determined by gas chromatography using a Shimadzu® GC-2010 Plus chromatograph equipped with AOC-20i automatic injector, Stabilwax-DA™ capillary column (30m, 0.25mm ID, 0.25 μ m df, Restek®) and flame ionization detector (FID).

The experimental design was a 2x2 factorial randomized block design, with 2 cutting regimes (0 and 1 cut) and 2 application management (with and without fungicide), with six replications, where each replication is represented by a plot of 9.45m².

Data were subjected to Shapiro-Wilk and Bartlett tests to check the assumptions of normality and homogeneity of variance, respectively. Once these assumptions were met, the F-test was applied at 5% confidence probability, by means of analysis of variance (ANOVA) at 5% significance level in SAS software (SAS Institute [SAS], 1993).

Results and Discussion

The results of the analysis of variance indicate significant interaction ($P < 0.05$) between the factors of cut management system and foliar application of fungicide for the parameters acetic acid and butyric acid, the other qualitative parameters of rye silages showed no interaction between the study factors.

The dry matter content (Table 1) was higher ($P < 0.05$) in the treatment subjected to a cut at the vegetative stage and after ensiling with an average of 49.12% DM, when compared to the treatment without cut at the vegetative stage, with 45.04% DM. Regarding DM losses, it was not possible to detect significant differences between the cutting systems, with an average of 4.63%. The pH was higher ($P < 0.05$) in the silages made of rye subjected to a cut at the vegetative stage, with an average of 4.33, compared to the treatment without cut, with 4.12. Lehmen, Fontaneli, Fontaneli and Santos (2014) analyzed different winter cereals without cut at the vegetative stage, including Rye cv. BRS Serrano with 33.69% dry matter content, and reported pH values that corroborate the present study, 4.26, and dry matter losses during ensiling of 3.3%.

Table 1
Fermentation and chemical characteristics of cv. Temprano rye silages subjected to cutting systems with or without fungicide leaf application at pre-flowering

Cutting system	Fungicide management		Mean
	With	Without	
Dry matter, %			
No cut	44.78	45.30	45.04 B
One cut	46.94	51.31	49.12 A
Mean	45.86 a	48.30 b	47.08
DM losses, % DM			
No cut	2.48	5.13	3.81 A
One cut	4.60	6.32	5.46 A
Mean	3.54 a	5.73 b	4.63
pH			
No cut	4.14	4.10	4.12 B
One cut	4.30	4.35	4.33 A
Mean	4.22 a	4.23 a	4.22

Means, followed by different uppercase letters in the same column, when comparing cutting systems, are significantly different by F-test at 5% significance.

Means, followed by different lowercase letters in the same row, when comparing fungicide managements, are significantly different by F-test at 5% significance.

Van Soest (1994) suggests that the higher the silage DM content, the pH is no longer an efficient parameter for silage quality evaluation, since the low moisture and high osmotic pressure inhibit the acidification of the medium. This leads to an inverse relationship between pH and moisture, where pH no longer necessarily indicates lower quality silage.

The use of fungicide at pre-flowering determined lower ($P < 0.05$) dry matter content in silages with average of 45.86% compared to the treatment without the use of fungicide with average of 48.30%, the same behavior was observed for DM losses during the fermentation process, with averages of 3.54% against 5.73% when no fungicide was applied, which corresponds to a 61.78% increase in dry matter losses. This is related to the physiological action on Pyraclostrobin in the plant, which according to Fagan et al. (2010), lead to a reduction in ethylene production, thereby delaying the senescence of leaves, thus increasing

the photosynthetic capacity of plants, also called “green effect”, justifying the lower DM content. This resulted in a better fermentation process and generated lower losses of DM, however the pH did not differ significantly with the use of fungicide, with an average of 4.22.

According to Muck (1988), this lower DM content, in turn, is closer to the ideal value for the fermentation process, which is 30% to 35%, since water activity is important for the microorganisms present in plants to turn soluble carbohydrates into organic acids, leading to an expected drop in pH below five, thus avoiding undesirable fermentations, in addition to the reduction of proteolysis that is inevitable in the process, which justifies lower losses with the use of fungicide.

The silage CP content (Table 2) was not different ($P > 0.05$) between the cutting systems, with an average of 6.99%, the same behavior was observed with HEM, with an average of 15.49%. The values

of the other parameters evaluated were significantly higher in the treatment without cut before ensiling, compared to the one subjected to a cut and then ensiling, among them MM (3.95% versus 3.53%), NDF (67.18% versus 62.64%), ADF (51.93% versus 46.92%), CEL (40.95% versus 37.82%) and LIG (10.98% versus 9.10%).

Table 2
Chemical composition of cv. Temprano rye silages subjected to cutting systems with or without fungicide leaf application at pre-flowering

Cutting system	Fungicide management		Mean
	With	Without	
Crude protein, % DM			
No cut	7.15	6.92	7.04 A
One cut	6.92	6.97	6.94 A
Mean	7.04 a	6.94 a	6.99
Mineral matter, % DM			
No cut	3.97	3.93	3.95 A
One cut	3.65	3.40	3.53 B
Mean	3.81 a	3.67 a	3.74
Neutral detergent fiber, % DM			
No cut	66.80	67.57	67.18 A
One cut	60.27	65.01	62.64 B
Mean	63.53 b	66.29 a	64.91
Acid detergent fiber, % DM			
No cut	51.75	52.11	51.93 A
One cut	44.80	49.03	46.92 B
Mean	48.28 a	50.57 a	49.42
Hemicellulose, % DM			
No cut	15.05	15.46	15.25 A
One cut	15.47	15.98	15.72 A
Mean	15.26 a	15.72 a	15.49
Cellulose, % DM			
No cut	41.00	40.91	40.95 A
One cut	36.76	38.88	37.82 B
Mean	38.88 a	39.89 a	39.39
Lignin, % DM			
No cut	10.75	11.20	10.98 A
One cut	8.04	10.15	9.10 B
Mean	9.39 b	10.68 a	10.04

Means, followed by different uppercase letters in the same column, when comparing cutting systems, are significantly different by F-test at 5% significance.

Means, followed by different lowercase letters in the same row, when comparing fungicide managements, are significantly different by F-test at 5% significance.

Fontaneli et al. (2009) evaluated different winter cereal silages including Rye silage, which presented CP values close to the present study, averaging 30% CP, but the fiber portions were lower: 59.80% NDF, 36.90% ADF. In the case of CP, Rotz and Muck (1994) observed similar values when silages were made at the same phenological stage, 7.20%.

Horst et al. (2018) examined silages of different winter cereals at the pre-flowering stage, comparing the rye cv. Temprano and cv. BRS Serrano, and found CP and MM values higher than those of the present study, 9.09% and 9.14% CP, respectively, for each material and 5.19% and 5.46% MM, respectively. In the same study, however, the NDF and ADF values were more similar to those found, with 73.93% and 45.95% respectively for cv. Temprano and 71.99% and 46.98% for cv. BRS Serrano. Neumann et al. (2019), on BR1 rye in the same field and at the same stage for silage making, observed similar values of MM, CP, NDF and ADF in the silage with averages of 3.56%, 7.05%, 57.07% and 38.02%, respectively.

The use of fungicide at pre-flowering led to a significant decrease in NDF, with values of 63.53% versus 66.29% of the control treatment, the same was observed with LIG, which presented 9.39% against 10.68%. As already mentioned, NDF is directly related to the ingestion capacity, allowing a higher consumption with the use of fungicides and a decrease in lignin allowing a better use of the food. The other parameters evaluated showed no differences ($P > 0.05$) with the use of fungicide, with averages of 6.99% CP, 3.74% MM, 49.42% ADF, 15.49% HEM and 39.39% CEL.

Venancio et al. (2019) analyzed wheat and black oats in silages at the same phenological stage and in the same experimental field, but with two applications, and also observed the same behavior with lower NDF values with fungicide application. Neumann et al. (2019) suggested that these differences in NDF and not found in ADF are due to the action of organic acids present during the fermentation process of silage, which may have led to hydrolysis of the hemicellulose fraction.

Ruminal disappearance of dry matter from rye cv. Temprano silage at 24 and 48 hours (Table 3) was not affected ($P > 0.05$) by different cutting systems or by the use of fungicide at pre-flowering, with averages of 41.44% and 51.07% respectively at the evaluated times. This was not expected because the treatment subjected to one cut showed better fiber index, which is the least digestible portion of the food, however, due to the better fermentation process and the evaluation was only 48 h, it was not possible the expression of the significant difference in quality.

In an experiment with rye silage at the same phase, Rotz and Muck (1994) found 54.2% *in vitro* digestibility, showing the genetic improvement of the material over the years, favoring its digestion. Horst et al. (2017) worked haylage production at the pre-flowering stage of cv. Temprano and BRS Sereno and found higher values of ruminal disappearance, 49.59% and 51.77% respectively in 24 h and 69.36% and 71.20% in 48 h, respectively, for each cultivar.

The behavior of the main organic acids and ethanol generated during the fermentation process (Table 3) was different; there was a significant interaction between the cutting system and fungicide management for acetic acid and butyric acid. The lactic acid was higher ($P < 0.05$) in the treatment without cut at the vegetative stage (32.08g kg⁻¹ DM) in relation to the treatment subjected to a cut (20.93g kg⁻¹ DM). Propionic acid and ethanol did not present significant differences in the different cutting systems with averages of 0.81g kg⁻¹ DM and 1.73g kg⁻¹ DM, respectively.

Acetic acid was lower ($P < 0.05$) within the interaction of factors in the treatment subjected to a cut without application of fungicide, with an average of 2.76 g kg⁻¹ DM, compared to the treatments subjected to a cut with fungicide, 7.33 g kg⁻¹ DM and treatments without cut that presented 6.80 g kg⁻¹ DM and 8.07 g kg⁻¹ DM with and without fungicide, respectively.

Table 3
Fermentation profile (g kg⁻¹) of cv. Temprano rye silages subjected to cutting systems with or without fungicide leaf application at pre-flowering

Cutting system	Fungicide management		Mean
	With	Without	
Lactic acid, g kg ⁻¹ DM			
No cut	32.06	32.10	32.08 A
One cut	23.30	18.56	20.93 B
Mean	27.68 a	25.33 a	26.50
Acetic acid, g kg ⁻¹ DM*			
No cut	6.80 A	8.07 A	7.43
One cut	7.33 A	2.76 B	5.04
Mean	7.06	5.42	6.24
Propionic acid, g kg ⁻¹ DM			
No cut	0.21	1.09	0.65 A
One cut	0.41	1.52	0.97 A
Mean	0.31 b	1.31 a	0.81
Butyric acid, g kg ⁻¹ DM*			
No cut	2.04 B	1.68 B	1.86
One cut	3.69 A	1.41 B	2.55
Mean	2.87	1.55	2.21
Ethanol, g kg ⁻¹ DM			
No cut	1.76	1.44	1.60 A
One cut	2.37	1.35	1.86 A
Mean	2.07 a	1.39 a	1.73

Means, followed by different uppercase letters in the same column, when comparing cutting systems, are significantly different by F-test at 5% significance.

Means, followed by different lowercase letters in the same row, when comparing fungicide managements, are significantly different by F-test at 5% significance.

* Means, followed by different uppercase letters in the columns and rows, are significantly different by Tukey's test at 5% significance in the interaction of cutting systems and fungicide managements.

Butyric acid was higher ($P < 0.05$) in the interaction in the treatment subjected to a cut with fungicide, with an average of 3.69 g kg⁻¹ DM compared to the others, without fungicide, 1.41 g kg⁻¹ DM and, the treatments without cut, averaging 2.04 g kg⁻¹ DM and 1.68 g kg⁻¹ DM, respectively, with and without fungicide.

J. S. Oliveira, Lanes, Lopes, Almeida and Carmo (2010) analyzed Triticale silages at different harvest dates, and reported values of acetic acid ranging from 19.3 to 70.5 g kg⁻¹ DM, propionic acid from

0.0 to 6.3 g kg⁻¹ DM, butyric acid from 0.8 to 4.5 g kg⁻¹ DM and finally lactic acid ranging from 9.7 to 120.5 g kg⁻¹ DM, thus showing a great variation in the results, as in the present study.

In a study carried out with BRS Umbu Wheat silage without cut, M. R. Oliveira, Bueno, Leão, Neumann and Jobim (2018) found values, in general, higher than the present study, Acetic acid 19.1 g kg⁻¹ DM, Propionic acid 5.2 g kg⁻¹ DM, Butyric acid 5.6 g kg⁻¹ DM, Lactic acid 17.6 g kg⁻¹ DM and Ethanol 7.7 g kg⁻¹ DM, results explained

by the greater participation of grains at the time of wheat ensiling.

The expected behavior during the preservation of forage as silage is that in the absence of oxygen there will be rapid pH decrease and stabilization, and consequently better preservation of the ensiled material. This drop directly depends on the amount of readily fermentable sugars present in the ensiled material and converted to organic acids. If it is appropriate, the conditions are more favorable for the development of lactic acid bacteria species present in the medium, however, the type of fermentation may be different and vary mainly by the different concentrations of acetic and lactic acids (Tabacco, Piano, Revello-Chion, & Borreani, 2011).

High oxygen concentrations, if there are delays in closing the silo or low compaction of the ensiled mass, will lead to a prolongation of plant respiration and predominance of bacteria of the genus *Clostridium*, as well as fungi and yeasts, thereby reducing lactic acid bacteria, which decreases the amount of acid leading to a lower pH drop and causing the temperature to rise (McDonald, Henderson, & Heron, 1991).

Sá, Nussio, Zopollatto, Junges and Bispo (2013) reported that high concentrations of butyric acid indicate the action of enterobacteria during the fermentation process and such a risk, if the process is poorly done, is higher in silages with increased nutrient availability for development of these spoilage microorganisms.

In agreement with Cherney and Cherney (2003), neutral products can be generated during the fermentation process, including ethanol, propanol,

and 2,3 propanediol, among others; the ethanol concentration can vary from 10 to 50 g kg⁻¹ DM and, as long as a period of adaptation of the animals is allowed, it has no complications and can be converted to acetate in the rumen environment.

The use of fungicide did not significantly alter the concentrations of lactic acid and ethanol in silages with average of 26.50 g kg⁻¹ DM and 1.73 g kg⁻¹ DM, respectively. Propionic acid in turn was lower (P <0.05) with the use of fungicide with an average of 0.31 g kg⁻¹ DM with 1.31 g kg⁻¹ DM when used, indicating that a change may have occurred in the fermentation process, regarding this specific organic acid, with the fungicide.

The silage maximum temperature after opening the silo (Table 4) was higher (P <0.05) in the treatment without cut at the vegetative stage after 216h compared to the treatment subjected to a cut after 176 h. The loss of aerobic stability also showed the same behavior being higher (P <0.05) in hours in the treatment without cut 240h in relation to the treatment subjected to a cut in 216h after opening the silo. This can be explained by the better fermentation process in the treatment without cut, which preserved higher nutrient content and consequently resulting in more substrate for proliferation of aerobic organisms, such as fungi and yeast (McDonald et al., 1991).

M. R. Oliveira et al. (2018) reported loss of stability with 141.3h after opening the silo. Neumann et al. (2019), in turn, obtained values of 160h for loss of stability in BR1 rye silages, not differing from other cereals such as BRS Cauê barley and CD 1440 wheat with the same 160h.

Table 4
Time to maximum temperature after opening and loss of aerobic stability of cv. Temprano rye silages subjected to cutting systems with or without fungicide leaf application at pre-flowering

Cutting system	Fungicide management		Mean
	With	With	
	Time to maximum temperature, hours		
No cut	216	216	216 A
One cut	176	176	176 B
Mean	196	196 a	
	Time to break aerobic stability, hours		
No cut	240	240	240 A
One cut	216	216	216 B
Mean	228 a	228 a	

Means, followed by different uppercase letters in the same column, when comparing cutting systems, are significantly different by F-test at 5% significance.

Means, followed by different lowercase letters in the same row, when comparing fungicide managements, are significantly different by F-test at 5% significance.

Aerobic stability is directly related to the pH and temperature of the material from contact with oxygen. Butyric acid-producing bacteria and yeast are susceptible to the acidic environment generated in fermentation, so low pH levels tend to lead to higher material preservation (Rodrigues et al., 2015). The temperature of the material is directly related to the activity of yeasts that develop in aerobic environment consuming the compounds formed in the fermentation, leading to deterioration and consequently pH increase. At this point, it is worth mentioning the acetic acid that plays an important role in the preservation of the material acting directly on the yeast and inhibiting its action (Kung Jr, Taylor, Lynch, & Neylon, 2003).

Muck (1988) mentions that the epiphytic flora of some winter cereals, composed of heterofermentative agents, causes higher concentrations of acetic acid 1,2 propanediol, which is transformed into propionic acid. In turn, Leão et al. (2017) assumed that the higher stability attributed to some winter cereal silages may be related to this higher production of organic acids with antifungal potential, thus avoiding temperature increase.

The use of a single fungicide application was not sufficient to significantly affect the parameters time to maximum temperature reached (196 h) and time to break aerobic stability (228 h), a fact that may be justified by the high disease pressure observed in all treatments. In an experiment with corn silage, the use of Pyraclostrobin altered its aerobic stability, Haerr, Lopes, Pereira, Fellows and Cardoso (2015) found higher aerobic stability of silage with three applications of Pyraclostrobin. The same authors presented higher concentrations of ethanol after fermentation in the treatment with Pyraclostrobin, probably due to the higher sugar concentration at harvest.

Silage aerobic stability breakdown indicates the onset of deterioration by microorganisms (Bernardes & Chizzotti, 2012). This proliferation of microorganism, mainly fungi, can produce mycotoxins, which generate nutritional losses and reduction of voluntary intake by animals. To avoid such a situation, McDonald et al. (1991) recommend the daily removal of an average 20 cm layer from the silo.

Conclusion

Silages of the treatments subjected to a single cut at the vegetative stage showed better characteristics in the general context, due to the better fiber composition with less lignification; however, due to its higher DM content, it presented a less efficient fermentation process. Nevertheless, the organic acid concentration and aerobic stability did not alter the DM losses or ruminal degradability, and there is also the use of DM harvested at the vegetative stage that certainly has a superior quality.

The use of fungicide provided only better dry matter index and better fiber composition and improved the fermentation process resulting in lower DM losses without altering the aerobic stability of silages. However, studies with more applications of fungicides should be carried out to perhaps further improve these indices.

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