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Effect of grain maturity stage on the quality of sorghum BRS-610 silages

[Efeito do estádio de maturação dos grãos na qualidade das silagens do sorgo BRS-610]

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ABSTRACT

The quality of sorghum BRS-610 silages in seven stages of grain maturity, between the milky and dry stage, for determination of the ideal moment of ensilage was evaluated. The silos were open after 56 days of fermentation. The values for dry matter of silages increased from 199 to 473g/kg or from 19.9 to 47.3%, with the advance of stage maturity of grains. As for the quality of fermentation, the silages may be classified as excellent quality in every stage of maturity. The lower values of neutral detergent fiber, 539g/kg or 53.9%, and acid detergent fiber, 307g/kg or 30.7%, were observed between the milky/dough to dough/dent stages. Regression analysis indicated an increase in values of lignin – 0.3 unit por stage – and reduction in values of *in vitro* dry matter digestibility – -2.5 units por stage – with the advance of the grain's stage maturity. The ensilage of sorghum BRS-610 between the milky/dough and dough stage ensure very good fermentation and nutritive value of silage.

Keywords: nutritive value, classification, digestibility, harvest moment, sorghum silage

RESUMO

Avaliou-se a qualidade das silagens do sorgo BRS-610 em sete estádios de maturação dos grãos, entre o estádio leitoso a estádio seco, para determinar o momento ideal de ensilagem. Os silos foram abertos após 56 dias de fermentação. Os teores de matéria seca das silagens aumentaram de 199 para 473g/kg, ou de 19,9 para 47,3%, com o avanço do estádio de maturação dos grãos. Quanto à qualidade de fermentação, as silagens foram classificadas como de excelente qualidade para todos os estádios de maturação. Os baixos valores de fibra detergente neutro, 539g/kg ou 53,9%, e fibra detergente ácido, 307g/kg ou 30,7%, foram observados entre os estádios leitoso/pastoso a pastoso/farináceo. A análise de regressão indicou aumento nos teores de lignina, 0,35 unidades por estádio, e redução nos valores de digestibilidade in vitro da matéria seca, -2,50 unidades por estádios de grãos leitoso/pastoso a pastoso, garante excelente fermentação e bom valor nutricional da silagem.

Palavras-chave: silagem de sorgo, classificação, digestibilidade, momento colheita, valor nutritivo

INTRODUCTION

Ensilage is one of the best forms of forage conservation used in Brazil, ensuring feed for ruminants during the dry season. According to McDonald *et al.* (1991), adequate forage preservation through the ensilage process depends on the production of lactic acid and on maintaining low pH values, as well as on low concentrations of ammoniacal nitrogen and acetic and butyric acids. The literature suggests

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that good quality sorghum ensilage may be obtained with amounts of dry matter varying from 20% to 35% (Borges *et al.*, 1997; Bernardino *et al.*, 1997; Brito *et al.*, 2000; Rocha Jr. *et al*, 2000; Pires *et al.*, 2006).

The maturity stage may influence the chemical composition and alter conservation quality as well as the nutritional value of sorghum silages. Sorghum hybrids present divergent behavior which is mainly associated to the difference in proportions between the plant's structural components. Differences in the amounts of neutral detergent fiber, acid detergent fiber, lignin and in vitro dry matter digestibility were not observed by Araújo et al. (2007) for hybrids of double purpose harvested between the milky and dry stages. On the other hand, different behavior between the hybrids for these variables was observed by Pires et al. (2006) while evaluating eight stages of maturity (from flowering to dry grain) of double purpose hvbrids.

BRS-160 Sorghum presents dry stalk, without tannin, and was obtained from the cross between CMSXS 232A and CMSXS 234R sorghums, from the sorghum breeding program at Embrapa Corn and Sorghum. BRS-160 stands out for its resistance to pests and diseases, good adaptability to the *cerrado* due to its tolerance to acid soils and drought, good resistance to lodging, which favors mechanical harvesting, with minimal losses in the field (Rodrigues et al., 2004). However, there is little information available concerning the quality of this hybrid's silage between the plant's stages of maturity. This is primordial for determining the adequate interval or maturity stage for ensilage of this hybrid, which will ensure good conservation of the silage forage and good nutritional value.

The objective of this study was to evaluate the quality of sorghum BRS-610 (*Sorghum bicolor* (L.) Moench) silages, in seven stages of maturity, from the milky stage to the dry grain stage.

MATERIALS AND METHODS

Sorghum BRS-610 was cultivated, harvested and ensiled at the National Research Center of Corn and Sorghum – Embrapa, in Sete Lagoas, MG, during the agricultural year of 2005/2006. The material was planted in 32 beds (7m vs 3.5m), with row spacing of 70cm. Cutting was done at soil level in the 2 central rows, discarding one meter on the extremities of each bed. The sorghum was planted on November 26th, 2005, with milky stages being reached on March 2nd, 2006, 96 days after planting. Silage was produced in a seven-day interval starting on this date and corresponded to the following stages of grain maturity: milky, milky/dough, dough, dough/dent, dent, hard and dry.

The material was chopped, sampled homogeneously and stored in PVC laboratory silos (10cm diameter x 40cm height) .The silos were opened on the 56^{th} day of fermentation.

The ensilage was removed from the silos and homogenized. Part of the material was pressed and the silage extract obtained was immediately evaluated for pH and ammoniacal nitrogen relative to total nitrogen (N-NH₃/NT) by distillation with calcium chloride and magnesium oxide (Official..., 2000). Part of the silage extract (10mL) was frozen with metaphosphoric acid at a 5:1 dilution for posterior determination of organic acids in gas chromatography (chromatograph Shimadzu GC-17^a) using a Nukol capillary column (10m in length and ¹/₄ inch in diameter) packed with Cromosorb 101 (80–100 mesh) as the stationary phase.

The other part was weighed and pre-dried in an air-forced oven at 55°C for 72 hours for the determination of pre-dried matter. The pre-dried samples were milled at 1mm. Dry matter (DM) content was determined in an oven at 105°C and crude protein (CP) was determined by the Kjeldhal method (Official..., 2000). Neutral detergent fiber (NDF), acid detergent fiber (ADF) and lignin were determined by solubilization of cellulose with sulphuric acid (72% p/p), through the sequential method of Van Soest et al. (1991), using thermo-stable α amilase (Termamyl 120L), without sodium sulfite, according to Mertens, (2002), with the Fiber analyzer ANKOM²²⁰. The in vitro dry matter digestibility (IVDMD), was obtained according to Holden (1999) for the utilization of rumen simulator Daisy^{II} ANKOM[®]. The inoculo was obtained from a fistulated bovine maintained on a diet of Tifton grass 85 hay, ad libidum and a daily 3kg of commercial concentrate (20% CP), fed at 8am and 5pm. The ruminal liquid was

collected before feeding, from the liquid:solid interface region in the ruminal environment.

The experimental design used was completely randomized with four beds per stage of maturity. The data was submitted to a variance analysis. The averages were compared by the Scott-Knott grouping method, at 5% probability. The response curves as a function of maturity were obtained by regression analysis, using the F test. The SAEG statistical package was used (Sistema..., 2007). Silage fermentation quality was determined according to the point criteria proposed by Tomich *et al.* (2003) (Table 1).

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Table I	Classification	of silages	according to	the	noint criteria
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pH values associated with dry matter content in the silage								
Dry matter content (%)								
	<20	20-30	30-40	>40	Point			
	<4.0	<4.2	<4.4	<4.6	25			
	>4.0-4.2	>4.2-4.4	>4.4-4.6	>4.6-4.8	20			
pH values	>4.2-4.4	>4.4-4.6	>4.6-4.8	>4.8-5.0	15			
	>4.4-4.6	>4.6-4.8	>4.8-5.0	>5.0-5.2	10			
	>4.6-4.8	>4.8-5.0	>5.0-5.2	>5.2-5.4	5			
	>4.8	>5.0	>5.2	>5.4	0			
N-NH ₃ /N	NT (%)	Butyric acid (%MS)		Acetic acid (%MS)				
Value	Point	Value	Point	Value	Point			
< 10.0	25	0.0-0.1	50	<2.5	0			
10.0 - 13.0	20	>0.1-0.3	40	40 >2.5-4.0				
> 13.0 - 17.0	15	>0.3-0.5	30	>4.0-5.5	-10			
> 17.0 - 21.0	10	>0.5-0.7	20	>5.5-7.0	-15			
> 21.0 - 25.0	5	>0.7-0.9	10	>7.0-8.5	-20			
> 25.0	0	>0.9	0	>8.5	-25			
Final Classification								
Final Score	Excellent 90-100	Good 70-89	Fair 50-69	Bad 30-49	Very bad <30			

Compiled from Tomich et al. (2003)

RESULTS AND DISCUSSION

The values for silage DM increased with the advancement of the grain maturity stage, however, the evaluation of the averages has shown significant differences only in the transition from dough to dough/dent stages and between the hard and dry stages (Table 2).

Regression analysis indicated linear increases (P<0.01) of 3.7 units in the amounts of DM in the silage between the stages, which were in one week intervals (Figure 1).

Grain maturity stage									
Variable	Milky	Milky/Dough	Dough	Dough/Dent	Dent	Hard	Dry	CV(%)	
DM (%)	19.86c	24.15c	24.48c	27.59b	30.52b	32.37b	47.31a	11.98	
pН	3.7d	3.8d	3.9c	4.0c	4.1b	4.2b	4.5a	2.90	
N- NH ₃ /NT ¹	1.25d	1.44d	2.26c	2.54c	4.26b	4.42b	5.99a	22.30	
Lactic acid ²	10.48a	8.66a	7.64a	7.92a	7.56a	5.16b	3.80b	21.26	
Acetic acid ²	2.72a	1.90a	2.39a	2.72a	2.65a	1.95a	0.89b	26.97	
Butyric acid ²	0.03c	0.03c	0.03c	0.02c	0.07a	0.04b	0.12a	38.16	

Table 2. Fermentative parameters for the silage of sorghum BRS-610 produced in seven stages of grain maturity

Means within a row without a common letter differ (P<0.05) by Scott-Knott test. ¹ % of ammoniacal nitrogen in relation to total nitrogen; ² % dry matter.

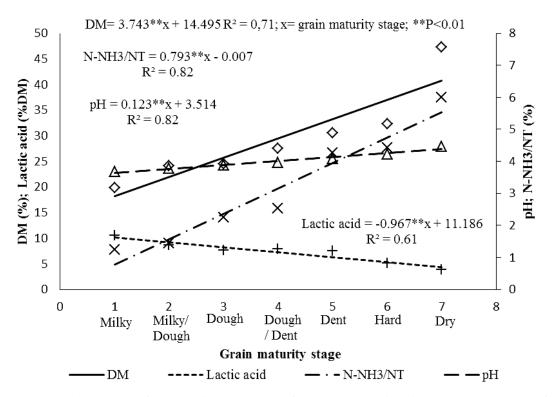


Figure 1. Tendency curves for the estimated values of dry matter, lactic acid, N-NH₃/NT and pH for silage of sorghum BRS-610 in seven stages of grain maturity. 1-milky; 2-milky/dough; 3-dough; 4-dough/dent; 5-dent; 6-hard; 7-dry; with one week intervals between stages, starting with flowering (0).

Araújo *et al.* (2007) observed DM increases ranging from 4.0 to 5.4 units between stages of maturity of double purpose sorghum silages with the advancement of the maturity stage. Molina *et al.* (2002) and Pires *et al.* (2006) also observed an increase in DM amounts with the advancement of the stage of maturity, associated to an increase in the participation of the sorghum plant's panicle. The amounts of DM necessary to ensure good conservation of the silage must be evaluated along with the pH values, because even silages with lower amounts of DM can be well preserved as long as they present low pH values, as suggested by Tomich *et al.* (2003). Values for the silages pH increased with the advancement of grain maturity stage (Table 2), however, the values obtained seem to have guaranteed the good conservation of the silages. Increases in pH values with maturity were observed by Pires *et al.* (2006) and Araújo *et al.* (2007). According to these authors the elevation in the amounts of DM are responsible for the elevation in the pH values, presenting high correlations (r = 0.82 and r = 0.86, respectively, P<0.01) among the variables.

Values for N-NH₃/NT were very low (Table 2), but increased linearly in the order of 0.8 units per stage of maturity (Figure 1). Pires *et al.* (2006) observed higher values for N-NH₃/NT (6.0% a 7.8%), which then reduced from flowering to the stage of dry grain, due to the elevation of the amount of DM with maturity. On the other hand, alterations in the levels of N-NH₃/NT with maturity were not observed by Araújo *et al.* (2007). These authors observed average levels of N-NH₃/NT of 5.5%; 5.4% and 7.1% for the sorghum hybrids BR700, BR701 and Massa-3, respectively, in PVC experimental silos.

Variations for this parameter are a reflex of the falling speed of pH, which inhibits the activity of proteolytic enzymes from the plant, as well as from the proteolytic microorganisms present in the silage. The elevation in the DM levels, pH and the reduction of lactic acid levels may be responsible for the elevation in the levels of N-NH₃/NT, due to some degree of proteolysis by the plant enzymes in the more advanced stages of maturity. Very good quality silages present levels of N-NH₃/NT between 0-10%. Therefore, the silage from hybrid BRS-610 may be considered of very high quality for all stages of maturity. This suggests that the proteolysis which took place during the fermentative process within the silo was reduced, thus preserving the quality of the ensiled forage.

The organic acids most commonly determined in the silages were lactic, acetic and butyric acids. Values for lactic acid were not different until the dent stage, decreasing in aftermost stages (Table 2). However, regression analysis indicated a linear fall of 0.9 units at each stage of maturity. This behavior agrees with results from Moisio and Heikonen (1994) and Araújo *et al.* (2007) who reported reductions in lactic acid content as the levels of DM in the silage increased. Inferior values of lactic acid were observed by Rocha Jr. *et al.* (2000) (3.2% to 8.5%). Based on this parameter the silage produced until the hard grain stage can be considered of very high quality (levels of lactic acid above 5.0%), according to Borges *et al.* (1997).

The values for acetic acid were not different up to the dent grain stage, however, values higher than 2.5% were observed in the milky, milky/dent and dent stages (Table 2). Borges et al. (1997) and Araújo et al. (2007) found correlation of r = 0.79 and r = 0.63 (P<0.01), respectively, between acetic and lactic acids. These authors suggest that in these conditions, great part of the acetate is derived from heterofermentative fermentations. In this study there was high and positive correlation (P<0.01) between lactic and acetic acids (r = 0.74). Due to the low values of ammoniacal nitrogen and butyric acid, it is possible that a situation similar to that described by these authors may have occurred.

The values for butyric acid were constant and low until the dough/dent stage, however, for successive stages, significant increases were observed (Table 2). Although there were variations in butyric acid content the values were low, which suggests good forage conservation, and may imply that there was a limitation in the development of microorganisms from the genera *Clostridium sp.* Borges *et al.* (1997), Rocha Jr. *et al.* (2000) and Araújo *et al.* (2007) produced good quality silages with butyric acid values ranging from 0% to 0.3%.

As to the quality of fermentation, the silages may be classified as excellent for all the stages of maturity, reflecting the aptitude of the hybrid for conservation in the form of silage (Table 3).

The levels of CP presented little variation with the stage of maturity, with lower values (P<0.05) between the dough/dent and dent stages. The advancement of the maturity stage did not result in CP level reductions in the silage. On the other hand, reductions in the values of CP with the advancement of maturity were observed by Pires *et al.* (2006) and Araújo *et al.* (2007). Regression analysis did not indicate a model that was adequate to the response, showing low R^2 values and no significance.

grain maturity stage									
Variable	Milky	Milky/Dough	Dough	Dough/Dent	Dent	Hard	Dry		
MS/pH	25	25	25	25	25	25	25		
$N-NH_3/NT^2$	25	25	25	25	25	25	25		
Acetic acid	-5	0	0	-5	-5	0	0		
Butyric acid	50	50	50	50	50	50	40		
Score	95	100	100	95	95	100	90		
Classification				Excellent					

Table 3. Scores and classification for the silage of sorghum BRS-610 produced in seven stages of grain maturity¹

¹Proposed by Tomich *et al.* (2003). ²Ammoniacal nitrogen in relation to total nitrogen.

Table 4. Chemical composition and *in vitro* DM digestibility for sorghum BRS-610 silage produced in seven stages of grain maturity

Variable	grain maturity stage							
(%DM)	Milky	Milky/Dough	Dough	Dough/Dent	Dent	Hard	Dry	CV
СР	7.27a	6.93a	6.86a	6.24b	5.99b	6.77a	6.27a	5.84
NDF	58.85a	53.88b	54.66b	53.14b	58.20a	61.19a	58.43a	6.59
ADF	33.56a	30.57b	31.13b	30.40b	33.22a	35.89a	35.05b	7.21
Hemicelluloses	25.29a	23.31b	23.54b	22.74b	24.97a	25.31a	23.70a	5.40
Cellulose	28.35	25.15	25.95	24.16	27.22	29.75	27.01	8.38
Lignin	5.21b	5.42b	5.18b	6.24b	6.00b	6.00b	7.72a	13.00
IVDMD	60.52a	60.13a	59.77a	53.05b	50.57b	45.05c	49.75b	4.85

Means within a row without a common letter differ (P < 0.05) by Scott-Knott test. CP: Crude protein. NDF: Neutral detergent fiber. ADF: Acid detergent fiber. IVDMD: *in vitro* DM digestibility.

The levels of NDF, ADF and hemicelluloses presented similar trends and showed reductions between the milky/dough and dough/dent stages. The highest values for NDF, ADF and hemicelluloses observed in the milky stage may be associated to the low participation of the panicle. On the other hand, in the more advanced stages the increase in levels for these fractions in the leaf and stalk may have suppressed the effect of the higher participation of the panicle in the plant (Faria Jr., 2008).

Pires *et al.* (2006) observed reduction in the fractions of NDF and ADF with the advancement of the stage of maturity due to the increase in the panicle in the sorghum plant. On the other hand, alterations in the fractions of NDF, ADF and lignin between the milky and hard grain stage of maturity in sorghum silages were not observed by Araújo *et al.* (2007). Results in the literature for sorghum silage, ensiled in the milky to dough

stages indicate values for NDF and ADF ranging from 44.6% and 63.6% and 20.2% to 31.3%, respectively. Based on the results obtained for BRS-610 it is possible to infer that it presents good quality in the fibrous fraction, displaying fiber levels close to those reported for grain sorghums or double purpose sorghums. Regression analysis did not indicate a model that was adequate to the variables NDF, ADF, hemicelluloses and cellulose, showing low R² values and no significance.

Cellulose and lignin levels were not altered (P<0.05) by the stage of maturity except by the increase in lignin in the dry grain stage. However, regression analysis indicated increases (P<0.01) of 0.3 units in lignin levels at each grain maturity stage (Figure 2). Alterations in lignin levels with the advancement of stage of maturity were not observed by Araújo *et al.* (2007).

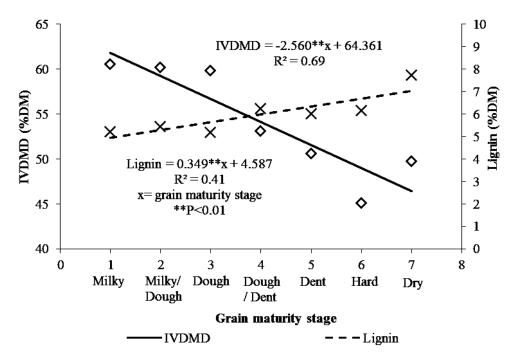


Figure 2. Tendency curves for the estimated values of *in vitro* DM digestibility (IVDMD) and lignin for silages of sorghum BRS-610 in seven stages of grain maturity. 1-milky; 2-milky/dough; 3-dough; 4-dough/dent; 5-dent; 6-hard; 7-dry; with one week intervals between stages, starting from flowering (0).

Pires et al. (2006) however, observed differences in the values for lignin between the hybrids related to the advancement in the stages of maturity. One hybrid presented an increase; another presented a decrease, while there were no alterations for this parameter in a third one. These authors reported lignin values ranging from 4.0% to 6.0%. However, even if the concentration of lignin does not alter its composition and the hydroxybenzaldehyde compounds ratio, vanillin and syringaldehyde may alter with the advancement of the stage of maturity (Noguera, 2002). According to Saliba et al. (1999), vanillin, which is derived from ferulic acid, is largely responsible for the bonds between lignin and hemicelluloses and for the reduction of the digestibility of forages.

The IVDMD levels were higher and similar among themselves in the early stage of maturity (milky to dough), with reduction for last stages (Table 4). However, regression analysis indicated increases (P<0.01) of 2.6 units in IVDMD at each grain maturity stage (Figure 2).

No effect of the maturity stage over IVDMD for the sorghum silages evaluated was found by Pires et al. (2006) and Araújo et al. (2007), with averages ranging from 54.9% to 57.8% and 47.2% to 52.2%, respectively. However, higher levels of IVDMD were observed by Silva et al. (1999) with the increase in the proportion of panicle in the ensiled mass. The effect of maturity stage over digestibility is variable in different materials due to the variation in the proportion of parts of the stalk, leaf and panicle in the plant and by the differences between the nutritional values of these fractions, which interferes in the final quality of sorghum silages. Based on the IVDMD values silages can be classified as good to average quality, according to Borges et al. (1997).

In the milky/dough to dough stages good nutritional values were observed, indicated by the lowest levels of fibrous fractions, good levels of digestibility and levels of dry matter which generally do not limit dry matter consumption by ruminants.

CONCLUSIONS

The grain maturity stage influenced both the quality of fermentation and the nutritional value of sorghum BRS-610 silages. Results from this experiment indicate that ensilage from sorghum BRS-610, between the milky/dough and dough stages, ensure the silage's high quality fermentation and good nutritional value.

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