



## Article

# Strategies to Maximize Kernel Processing in a Brazilian Vitreous Endosperm Hybrid

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**Abstract:** Whole-plant corn silage (WPCS) is a major source of forage for dairy cattle in Brazil. Improved kernel processing may be especially advantageous when feeding corn hybrids with vitreous endosperms. Two experiments were conducted to evaluate the effects of the theoretical length of cut (TLOC) and ensiling time on WPCS particle size and kernel processing with two types of forage harvesters. In the first trial, the plant was harvested by a pull-type forage harvester (PTFH) at TLOCs of 3, 6 and 9 mm. In the second trial, the harvesting was performed by a self-propelled forage harvester (SPFH) at TLOCs of 6, 12 and 18 mm, with a roll gap of 3 mm. The WPCS were stored for 0, 35 and 140 days. In the PTFH trial, the TLOCs of 3 and 6 mm did not affect the WPCS particle size distribution and mean particle length (MPL). However, the TLOC of 9 mm increased the MPL. The increase in the TLOC with the SPFH led to a higher MPL and percentage of long particles. The ensiling time increased the MPL and long particles only for the WPCS harvested by the SPFH. The strategy of reducing the TLOC with the SPFH increased the percentage of kernels smaller than 4.75 mm. Furthermore, the TLOC of 6 mm led to the best kernel processing with the SPFH. The ensiling time reduced the particle size of the kernel fractions for both forage harvesters. The corn silage processing score was improved with 140 days of ensiling with the SPFH. These findings suggest that increasing the ensiling time and a low TLOC in SPFHs (6 mm) may be good strategies to increase kernel damage and starch digestibility in WPCS.

**Keywords:** kernel particle size; vitreous endosperm; ensiling time; theoretical length of cut



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## 1. Introduction

Whole-plant corn silage (WPCS) is the predominant source of forage for lactating dairy cattle in Brazil [1] and worldwide [2]. WPCS is a high-energy forage composed of stover and kernel fractions, which are rich in neutral detergent fiber (NDF) and starch contents, respectively [3]. For the highest availability of starch in WPCS, the kernel processing is crucial [4]. Despite the increase in custom contractors offering self-propelled forage harvester (SPFH) services [5], a majority (90.4%) of Brazilian dairy farmers harvest their corn crops with a pull-type forage harvester (PTFH) without a kernel processor (KP) [1]. This is a concern because the most cultivated hybrids in Brazil have vitreous kernels with a higher vitreous endosperm proportion [6,7]. Furthermore, a higher vitreous endosperm is related to kernel hardness [6], which makes kernels more difficult to break down by the harvester's processors [2], and this could be worse for a PTFH without KP.

In WPCS processed by rollers during harvesting, the rollers fracture the corn kernels [8,9] and increase the total-tract starch digestibility (TTSD) [10]. Nonetheless, WPCS efficiency is also influenced by maturity, the theoretical length of cut (TLOC), the type of processor, roll speed differential and processor roll-gap clearance [4,9]. Kernel

vitreousness increases throughout the WPCS maturing process [8] and, with a high WPCS dry matter (DM) content (>40%), the TTSD is reduced [4]. This situation can occur in Brazilian vitreous corn hybrids, even with a DM below 40%.

The *in vitro* starch digestibility (ivSD) of WPCS is improved by a longer length of storage [11]. Extended time stored in silos leads to the proteolysis of zein proteins and disassociation of starch-protein clusters in high-moisture corn, which could allow for a greater surface area for rumen bacteria [12]. Higher values of proteolysis markers (soluble crude protein [CP] and ammonia-N), followed by an improvement in ivSD, have been observed in WPCS stored for long periods of time [13]. The corn silage processing score (CSPS) is a widely used method to assess the degree of kernel breakage and processing adequacy in WPCS. Ferraretto et al. [14] reported an increase of 7 to 10% for the CSPS in corn silage ensiled for at least 30 d and up to 240 d. Since CSPS is defined as the mass proportion of starch particles smaller than 4.75 mm, it is possible to presume that an increased length of ensiling could reduce kernel particle size, due to the proteolysis of the prolamin zein matrix, increasing the starch surface area for digestion [12].

Finally, our main goal is to provide practical recommendations that can be applied to the reality of the majority of dairy farms in Brazil, so the producers can take the best possible advantage of the harvesters they already have. In general, dairy farmers increase the TLOC with the aim of increasing the particle size of the WPCS and, thereby, achieving a greater physically effective fiber (peNDF) [2,15]. The TLOC is usually between 10 and 13 mm in unprocessed WPCS and about 19 mm in processed WPCS [16,17]. Ferraretto and Shaver et al. [4] reported no impact of kernel processing in ruminal starch digestibility when WPCS was harvested with a high TLOC (>32 mm). The hypothesis of our study is that, in order to achieve adequate kernel processing in a vitreous endosperm hybrid, a lower TLOC and a longer length of storage is required. The objective of this study was to evaluate the impact of two different types of forage harvesters, TLOC settings and ensiling time on the physical characteristics of WPCS.

## 2. Materials and Methods

### 2.1. Silage Production and Treatments

Three hectares of a vitreous corn hybrid (DKB 177 VT PRO 2, Dekalb, Monsanto, Dekalb, IL, USA) were sown in November 2015, at a density of 68,750 seeds per hectare at the Luiz de Queiroz College of Agriculture—University of Sao Paulo (Piracicaba, Sao Paulo, Brazil). The harvesting occurred in February 2016, achieving approximately 34.0% DM of whole-plant corn. The field was harvested in alternating strips in an attempt to keep similar nutrient compositions among the treatments. The treatments were performed during the harvest. The corn vitreousness was determined by manual dissection of the kernels [18] in fifteen ears collected from the corn crop, following the procedure described by Correa et al. [6].

In the first experiment, a JF AT 1600 PTFH without KP (JF Máquinas Agrícolas, Itapira, Brazil), set for TLOCs of 3, 6 and 9 mm, was used in the imposed harvesting treatments. Samples of 8.5 kg were ensiled in 10 L buckets (density of 850 kg m<sup>-3</sup>) and stored for 0, 35 and 140 d. The experiment consisted of nine treatments (three lengths of cut and three lengths of storage) in 36 mini-silos (four replications per treatment). In the second experiment, an FR 9050 SPFH (CNH Industrial, New Holland, PA, USA), equipped with conventional rollers set at a 3 mm roll gap and 20% roll speed differential, harvested the treatments with the following TLOCs: 6, 12 and 18 mm. All the samples were ensiled in the same manner as described in the first experiment. Again, this experiment also consisted of nine treatments (three lengths of cut and three lengths of storage) and 36 mini-silos (four replications per treatment). The mini-silos were stored in natural environmental conditions until achieving the targeted length of storage. After the storage period reached 35 d and 140 d, the mini-silos were opened and the samples were immediately manipulated.

## 2.2. Physical and Chemical Characteristics

The WPCS samples were homogenized manually and divided into four equal subsamples of 500 g using a quartering technique. The first subsample, the as-fed WPCS samples, was used for the determination of particle size distribution and MPL using a Penn State Particle Size Separator (PSPS). The PSPS procedure was conducted manually using three sieves (19, 8 and 1.18 mm) and a bottom pan, according to the method of Kononoff et al. [19].

The second subsample was dried at 60 °C for 48 h in a forced-air oven and then ground to pass through a 1 mm Wiley mill screen (Arthur H. Thomas). The DM (method 942.05) and ether extract (EE, method 2003.05) were determined as described by AOAC International [20]. The concentration of CP was determined as  $N \times 6.25$  after analysis with an N analyzer (Leco FP-2000; Leco Corp., St. Joseph, MI, USA) by the Dumas method [21]. The NDF was analyzed using a TE-149 fiber analyzer (TECNAL Equipamentos para Laboratórios, Piracicaba, Brazil) with heat-stable  $\alpha$ -amylase and sodium sulfite (method 2002.04) [20]. The starch was analyzed according to Hall et al. [22]. This data was used only for the description of the WPCS nutritional composition.

The third subsample was not ground. It was dried at 60 °C for 48 h in a forced-air oven to evaluate the corn silage processing score (CSPS), as described by Ferreira and Mertens [23]. The CSPS is defined as the mass proportion of starch particles smaller than 4.75 mm. In the fourth subsample, the kernel and stover fractions were separated by a hydrodynamic separation procedure [24]. This method is based on differences in buoyancy between the kernels and the stover. Because all the samples were fermented for at least 35 d, which makes the separation process more difficult, the samples were dried in a forced-air oven set at 60 °C for 48 h prior to immersion in water, as recommended by Savoie et al. [24]. Each sample of approximately 400 g of DM was placed in a 10 L rectangular tub containing 7 L of water. Each dried sample was gently agitated manually for 2 min, so that the entire sample was submerged in water. After 2 min, the stover fraction, which floated, due to a lower density than water, was removed gently using a small 1.18 mm sieve. After separation, the kernel fraction was transferred to aluminum plates, re-dried at 60 °C for 48 h in a forced-air oven and dry-sieved using a Tyler Ro-Tap Shaker (model RX-29, Tyler, Mentor, OH, USA) with a set of nine sieves, with nominal square apertures of 9.50, 6.70, 4.75, 3.35, 2.36, 1.70, 1.18 and 0.59 mm, and a bottom pan. The geometrical mean particle size (GMPS;  $\mu\text{m}$ ) and surface area ( $\text{cm}^2 \text{g}^{-1}$ ) were calculated using a log-normal distribution [25], as described by Dias Junior et al. [3]. The proportion of kernels smaller than 4.75 mm was calculated by the sum of the percentages retained on the 3.35, 2.36, 1.70, 1.18 and 0.59 mm sieves and bottom pan [3].

## 2.3. Statistical Analysis

The data from both trials were analyzed as a split-plot design using the procedure MIXED of SAS (SAS Institute Inc., Cary, NC, USA). The model included the fixed effects of the TLOC, ensiling time and the interaction of TLOC  $\times$  ensiling time. A mini-silo was used as the experimental unit. The means were determined using the LSMEANS statement and were compared using the Tukey test at 5% of significance. The statistical significance and trends were determined at  $p \leq 0.05$  and  $p > 0.06$  to  $p \leq 0.10$ , respectively.

## 3. Results

The kernel vitreousness of the hybrid was  $65.6 \pm 3.7\%$  of the total endosperm. The chemical composition of whole-plant corn is shown in Table 1. The mean and standard deviation per nutrient (% of DM) was  $6.4 \pm 1.4$  for CP,  $3.4 \pm 1.4$  for EE,  $43.6 \pm 0.8$  for NDF and  $30.6 \pm 1.7$  for starch. The average DM among the treatments (% of as-fed) was 35.2%.

**Table 1.** Chemical composition of unfermented whole-plant corn harvested with different forage harvesters at different theoretical lengths of cut settings.

Item <sup>1</sup>	PTFH <sup>2</sup>			SPFH <sup>3</sup>		
	3 mm	6 mm	9 mm	6 mm	12 mm	18 mm
DM,% of as-fed	34.2	33.9	34.0	34.5	34.3	34.1
CP	5.0	6.5	4.9	5.6	8.2	8.2
EE	3.2	3.0	3.1	3.2	3.7	4.2
NDF	42.2	43.9	44.1	39.7	44.0	44.5
Starch	30.6	28.2	32.8	32.5	28.8	30.6

<sup>1</sup> Items are presented as percentage of dry matter (% DM) unless otherwise noted. DM: dry matter; CP: crude protein; EE: ether extract; NDF: neutral detergent fiber. <sup>2</sup> PTFH: pull-type forage harvester. <sup>3</sup> SPFH: self-propelled forage harvester.

### 3.1. Experiment 1

Reducing the TLOC from 6 to 3 mm did not impact the particle size distribution and MPL in the PTFH samples (Table 2). However, the highest TLOC (9 mm) increased the percentage of particles retained on the top two PSPS sieves which, as a consequence, led to the decreased retention of material on the 1.18 mm sieve and bottom pan. A greater MPL (11.5 mm) was observed for this TLOC.

**Table 2.** Effect of theoretical length of cut on particle size distribution and mean particle length in whole-plant corn silage harvested with a pull-type forage harvester.

Item <sup>1</sup>	TLOC <sup>2</sup>			SEM <sup>3</sup>	<i>p</i> -Value
	3 mm	6 mm	9 mm		
Sieve					
19 mm	9.8 <sup>b</sup>	8.2 <sup>b</sup>	13.7 <sup>a</sup>	0.83	<0.01
8 mm	69.3 <sup>b</sup>	68.9 <sup>b</sup>	72.4 <sup>a</sup>	0.66	0.03
1.18 mm	18.9 <sup>a</sup>	20.8 <sup>a</sup>	12.6 <sup>b</sup>	0.61	<0.01
Bottom pan	1.9 <sup>a</sup>	1.9 <sup>a</sup>	1.4 <sup>b</sup>	0.10	0.01
MPL, mm	10.2 <sup>b</sup>	9.8 <sup>b</sup>	11.5 <sup>a</sup>	0.15	0.01
Sgm	1.96 <sup>a</sup>	1.95 <sup>a</sup>	1.86 <sup>b</sup>	0.153	<0.01

<sup>1</sup> Percentage of particles retained on each sieve (as-fed basis) of Penn State Particle Size Separator; MPL: mean particle length calculated, as described by Kononoff et al. [19]; Sgm: geometric standard deviation.

<sup>2</sup> TLOC: theoretical length of cut. <sup>3</sup> SEM: standard error of mean. <sup>a,b</sup> Means in the same row with different superscript letters differ by Tukey test ( $p \leq 0.05$ ).

The ensiling time did not change the particle size distribution and MPL (Table 3). There was no interaction between the TLOC and ensiling time for the particle size distribution results ( $p > 0.20$ ).

**Table 3.** Effect of ensiling time on particle size distribution and mean particle length in whole-plant corn silage harvested with a pull-type forage harvester.

Item <sup>1</sup>	Ensiling Time (Day)			SEM <sup>2</sup>	<i>p</i> -Value
	0 d	35 d	140 d		
Sieve					
19 mm	10.8	9.6	11.3	0.78	0.32
8 mm	70.1	71.0	69.7	0.75	0.46
1.18 mm	17.2	17.9	17.2	0.61	0.62
Bottom pan	1.8	1.6	1.8	0.10	0.12
MPL, mm	10.5	10.4	10.6	0.15	0.72
Sgm	1.93	1.90	1.95	0.15	0.12

<sup>1</sup> Percentage of particles retained on each sieve (as-fed basis) of Penn State Particle Size Separator; MPL: mean particle length calculated, as described by Kononoff et al. [19]; Sgm: geometric standard deviation.

<sup>2</sup> SEM: standard error of mean.

The different TLOCs did not change the kernel fraction particle size distribution (Table 4). As a result, the GMPS, surface area and proportion of kernel fraction passing through a 4.75 mm sieve were not affected among the treatments. The CSPA values were low and varied between 24.7 and 26.4%.

**Table 4.** Effect of theoretical length of cut on kernel fraction particle size distribution, geometric mean particle size, surface area and processing score in whole-plant corn silage harvested with a pull-type forage harvester.

Item	TLOC <sup>1</sup>			SEM <sup>2</sup>	p-Value
	3 mm	6 mm	9 mm		
Sieve <sup>3</sup> , mm					
6.7	36.3	35.9	36.9	0.97	0.79
4.75	38.4	39.3	38.8	0.81	0.79
3.35	8.8	8.8	8.9	0.28	0.95
2.36	6.3	6.3	6.2	0.36	0.98
1.70	3.2	3.1	2.9	0.16	0.58
1.18	2.3	2.1	2.1	0.12	0.30
0.59	2.6	2.5	2.4	0.14	0.57
Pan	2.1	2.1	1.9	0.13	0.51
% < 4.75 <sup>4</sup>	25.3	24.8	24.3	0.93	0.79
GMPS <sup>5</sup> , µm	5070.0	5090.4	5172.8	62.5	0.48
Surface area, cm <sup>2</sup> g <sup>-1</sup>	11.1	11.0	10.6	0.25	0.31
Processing score <sup>6</sup>	24.7	26.4	25.0	1.71	0.76

<sup>1</sup> TLOC: theoretical length of cut. <sup>2</sup> SEM: standard error of mean. <sup>3</sup> Percentage of particles retained on each sieve (DM basis). <sup>4</sup> Proportion of kernel fraction passing through a 4.75 mm sieve. <sup>5</sup> GMPS: geometric mean particle size. <sup>6</sup> Percent of starch passing through a 4.75 mm sieve, measured as described by Ferreira and Mertens [23].

Despite no effect from the TLOC, the ensiling time changed the kernel particle size distribution regardless of the extension of ensiling time, either 35 or 140 d (Table 5). The percentage of particles retained on the 6.7 mm sieve decreased by 13.2 and 11.3%, respectively, for 35 and 140 d of storage in comparison with fresh samples. There was an increase in particles retained on the 4.75, 1.18 and 0.59 mm sieves and pan. A higher surface area and proportion of kernels smaller than 4.75 mm were observed for both lengths of storage in comparison with the fresh samples. A reduction in GMPS occurred; however, the CSPA was not affected. No interaction was observed between the TLOC and ensiling time for the kernel fraction variables and CSPA ( $p > 0.11$ ).

### 3.2. Experiment 2

The WPCS particle size distribution was affected by the different TLOC settings on the SPFH samples (Table 6). The percentage of material retained on the top PPS sieve was different among the treatments (4.5, 8.0 and 15.6%, respectively, for the TLOC settings of 6, 12 and 18 mm). On the 8 mm sieve, the highest value (66.2% particles retained) was observed for the TLOC of 12 mm. For the TLOC settings of 6 and 18 mm, the amounts retained on the same sieve size were, respectively, 56.2 and 63.1%. The decrease in TLOC from 18 to 6 mm led to an increase in the percentage of material retained on the 1.18 mm sieve from 17.7 to 34.2%. The TLOCs of 12 and 18 mm resulted in minor amounts of particles in the PPS pan in comparison with the lowest TLOC (6 mm). An increase in the MPL occurred accordingly with the increments of TLOC.

**Table 5.** Effect of ensiling time on kernel fraction particle size distribution, geometric mean particle size, surface area and processing score in whole-plant corn silage harvested with a pull-type forage harvester.

Item	Ensiling Time (Day)			SEM <sup>5</sup>	p-Value
	0 d	35 d	140 d		
Sieve <sup>1</sup> , mm					
6.7	44.5 <sup>a</sup>	31.3 <sup>b</sup>	33.2 <sup>b</sup>	0.97	<0.01
4.75	32.9 <sup>b</sup>	42.9 <sup>a</sup>	40.7 <sup>a</sup>	0.81	<0.01
3.35	8.2 <sup>b</sup>	9.1 <sup>a</sup>	9.2 <sup>a</sup>	0.28	0.05
2.36	6.2	6.2	6.5	0.36	0.71
1.70	3.2 <sup>a</sup>	3.1 <sup>a</sup>	2.9 <sup>b</sup>	0.16	0.01
1.18	1.8 <sup>b</sup>	2.3 <sup>a</sup>	2.4 <sup>a</sup>	0.12	<0.01
0.59	2.1 <sup>b</sup>	2.7 <sup>a</sup>	2.7 <sup>a</sup>	0.14	0.02
Pan	1.6 <sup>b</sup>	2.4 <sup>a</sup>	2.3 <sup>a</sup>	0.13	<0.01
% < 4.75 <sup>2</sup>	22.3 <sup>b</sup>	25.8 <sup>a</sup>	26.5 <sup>a</sup>	0.84	0.03
GMPS <sup>3</sup> , µm	5419.1 <sup>a</sup>	4930.2 <sup>b</sup>	4980.3 <sup>b</sup>	62.5	<0.01
Surface area, cm <sup>2</sup> g <sup>-1</sup>	9.8 <sup>b</sup>	11.4 <sup>a</sup>	11.4 <sup>a</sup>	0.25	<0.01
Processing score <sup>4</sup>	26.1	23.4	26.6	1.65	0.37

<sup>1</sup> Percentage of particles retained on each sieve (DM basis). <sup>2</sup> Proportion of kernel fraction passing through a 4.75 mm sieve. <sup>3</sup> GMPS: geometric mean particle size. <sup>4</sup> Percent of starch passing through a 4.75 mm sieve, measured as described by Ferreira and Mertens [23]. <sup>5</sup> SEM: standard error of mean. <sup>a,b</sup> Means in the same row with different superscript letters differ by Tukey test ( $p \leq 0.05$ ).

**Table 6.** Effect of theoretical length of cut on particle size distribution and mean particle length in whole-plant corn silage harvested with a self-propelled forage harvester.

Item <sup>1</sup>	TLOC <sup>2</sup>			SEM <sup>3</sup>	p-Value
	6 mm	12 mm	18 mm		
Sieve					
19 mm	4.5 <sup>c</sup>	8.0 <sup>b</sup>	15.6 <sup>a</sup>	0.74	<0.01
8 mm	56.2 <sup>c</sup>	66.2 <sup>a</sup>	63.1 <sup>b</sup>	0.84	<0.01
1.18 mm	34.2 <sup>a</sup>	22.8 <sup>b</sup>	17.7 <sup>c</sup>	0.59	<0.01
Bottom pan	4.2 <sup>a</sup>	3.2 <sup>b</sup>	2.8 <sup>b</sup>	0.08	<0.01
MPL, mm	7.7 <sup>c</sup>	9.3 <sup>b</sup>	10.7 <sup>a</sup>	0.11	<0.01
Sgm	2.15 <sup>a</sup>	2.06 <sup>b</sup>	2.12 <sup>ab</sup>	0.016	<0.01

<sup>1</sup> Percentage of particles retained on each sieve (as-fed basis) of Penn State Particle Size Separator; MPL: mean particle length calculated, as described by Kononoff et al. [19]; Sgm: geometric standard deviation.

<sup>2</sup> TLOC: theoretical length of cut. <sup>3</sup> SEM: standard error of mean. <sup>a-c</sup> Means in the same row with different superscript letters differ by Tukey test ( $p \leq 0.05$ ).

The length of storage resulted in changes in the WPCS particle size distribution (Table 7). The longest ensiling time period evaluated (140 d) increased the number of particles in the 19 mm PSPS sieve in comparison with the other ensiling time periods. In the 8 mm sieve, both ensiling times (short and long) had higher percentages of particles retained than the unfermented WPCS. The MPL (7.9, 9.5 and 10.4 mm) increased along with the length of storage.

There was an interaction between the TLOC and ensiling time for the particles in the PSPS pan ( $p = 0.01$ ; Figure 1). Distinct amounts in the pan were observed at time 0 among the TLOC settings. Nonetheless, in the short and long ensiling time periods, there was an increase in the particles in the pan only for the 6 mm TLOC. For the other variables, no interactions occurred ( $p > 0.14$ ).

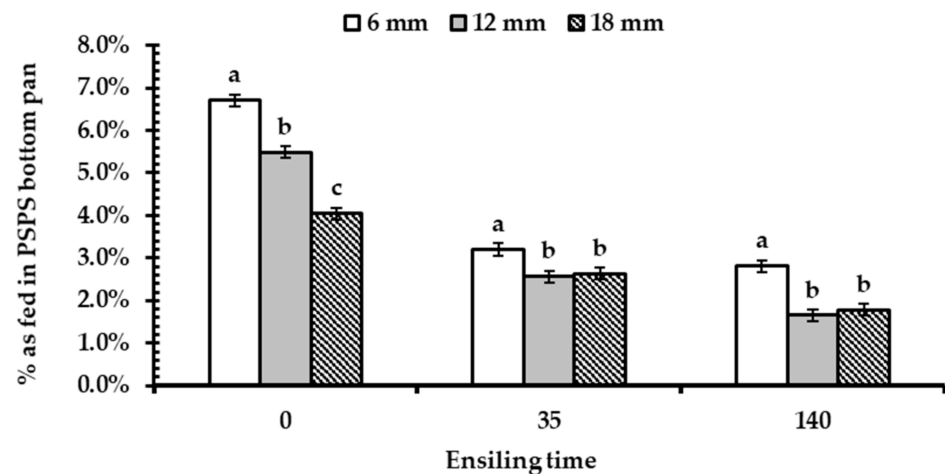


**Table 7.** Effect of ensiling time on particle size distribution and mean particle length in whole-plant corn silage harvested with a self-propelled forage harvester.

Item <sup>1</sup>	Ensiling Time (Day)			SEM <sup>2</sup>	<i>p</i> -Value
	0 d	35 d	140 d		
Sieve					
19 mm	7.8 <sup>b</sup>	7.8 <sup>b</sup>	11.7 <sup>a</sup>	0.74	<0.01
8 mm	54.0 <sup>b</sup>	66.1 <sup>a</sup>	65.4 <sup>a</sup>	0.84	<0.01
1.18 mm	31.5 <sup>a</sup>	22.4 <sup>b</sup>	20.8 <sup>c</sup>	0.59	<0.01
Bottom pan	5.4 <sup>a</sup>	2.8 <sup>b</sup>	2.1 <sup>b</sup>	0.08	<0.01
MPL, mm	7.9 <sup>c</sup>	9.5 <sup>b</sup>	10.4 <sup>a</sup>	0.11	<0.01
Sgm	2.28 <sup>a</sup>	2.05 <sup>b</sup>	2.00 <sup>b</sup>	0.016	<0.01

<sup>1</sup> Percentage of particles retained on each sieve (as-fed basis) of Penn State Particle Size Separator; MPL: mean particle length calculated, as described by Kononoff et al. [19]; Sgm: geometric standard deviation.

<sup>2</sup> SEM: standard error of mean. <sup>a–c</sup> Means in the same row with different superscript letters differ by Tukey test ( $p \leq 0.05$ ).

**Figure 1.** Interaction between theoretical length of cut and ensiling time (day) for particles in the Penn State Particle Size Separator bottom pan ( $p = 0.01$ ; SEM = 0.08) in whole-plant corn silage harvested with a self-propelled forage harvester. SEM: standard error of mean. <sup>a–c</sup> Means in the same ensiling time with different superscript letters differ by Tukey test ( $p \leq 0.05$ ).

The kernel particle size distribution changed through the TLOC settings (Table 8). In the upper sieve, the TLOCs of 6 (14.9%) and 12 mm (17.0%) retained lower percentages than the 18 mm (20.4%). In the next sieve (4.75 mm), there was an increase of 2.5% of particles retained in the 18 mm in relation to the 6 mm TLOC. However, the 12 mm was not distinguished from the other treatments. The TLOC affected the particle size distribution in the 3.35, 2.36, 1.70 and 1.18 mm sieves and pan differently. The kernels smaller than 4.75 mm increased (57.1, 60.8 and 65.0%) accordingly with the reduction in the TLOC. However, only the 6 mm TLOC affected the GMPs and surface area. The CSPs differed between the 6 and 18 mm TLOCs.

The impact of fermentation on kernel particle size distribution is shown in Table 9. In summary, both 35 and 140 d of ensiling reduced particles larger than 6.7 mm and increased the percentage of kernels retained in the sieves with apertures of 1.7, 1.18 and 0.59 mm.

**Table 8.** Effect of theoretical length of cut on kernel fraction particle size distribution, geometric mean particle size, surface area and processing score in whole-plant corn silage harvested with a self-propelled forage harvester.

Item	TLOC <sup>1</sup>			SEM <sup>2</sup>	p-Value
	6 mm	12 mm	18 mm		
Sieve <sup>3</sup> , mm					
6.7	14.9 <sup>b</sup>	17.0 <sup>b</sup>	20.4 <sup>a</sup>	0.78	<0.01
4.75	20.0 <sup>b</sup>	22.2 <sup>ab</sup>	22.5 <sup>a</sup>	0.66	0.03
3.35	13.2 <sup>c</sup>	15.8 <sup>a</sup>	14.3 <sup>b</sup>	0.23	<0.01
2.36	19.6 <sup>a</sup>	18.9 <sup>ab</sup>	17.6 <sup>b</sup>	0.40	<0.01
1.70	10.8 <sup>a</sup>	8.8 <sup>b</sup>	8.0 <sup>c</sup>	0.21	<0.01
1.18	7.1 <sup>a</sup>	5.3 <sup>b</sup>	5.3 <sup>b</sup>	0.24	<0.01
0.59	7.8 <sup>a</sup>	6.4 <sup>b</sup>	6.3 <sup>b</sup>	0.22	<0.01
Pan	6.5 <sup>a</sup>	5.3 <sup>b</sup>	5.4 <sup>b</sup>	0.21	<0.01
% < 4.75 <sup>4</sup>	65.0 <sup>a</sup>	60.8 <sup>b</sup>	57.1 <sup>c</sup>	1.01	<0.01
GMPS <sup>5</sup> , µm	2908.1 <sup>b</sup>	3206.7 <sup>a</sup>	3346.6 <sup>a</sup>	49.07	<0.01
Surface area, cm <sup>2</sup> g <sup>-1</sup>	20.0 <sup>a</sup>	18.5 <sup>b</sup>	18.1 <sup>b</sup>	0.27	<0.01
Processing score <sup>6</sup>	60.4 <sup>a</sup>	52.8 <sup>ab</sup>	51.1 <sup>b</sup>	2.37	0.02

<sup>1</sup> TLOC: theoretical length of cut. <sup>2</sup> SEM: standard error of mean. <sup>3</sup> Percentage of particles retained on each sieve (DM basis). <sup>4</sup> Proportion of kernel fraction passing through a 4.75 mm sieve. <sup>5</sup> GMPS: geometric mean particle size. <sup>6</sup> Percent of starch passing through a 4.75 mm sieve, measured as described by Ferreira and Mertens [23].  
<sup>a-c</sup> Means in the same row with different superscript letters differ by Tukey test ( $p \leq 0.05$ ).

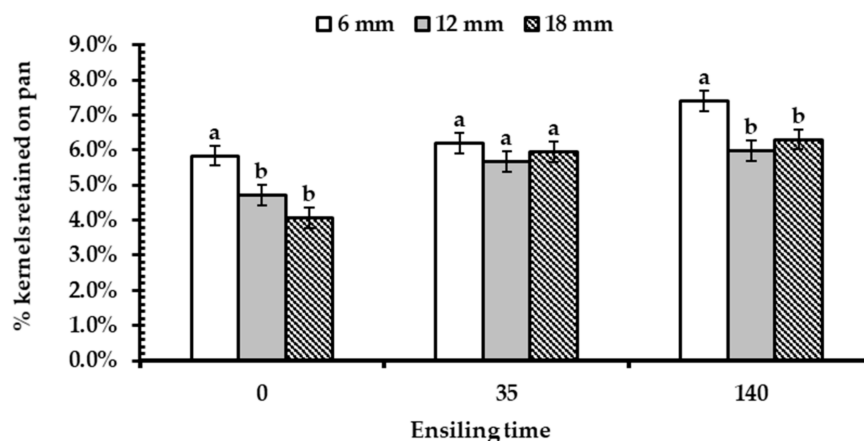
**Table 9.** Effect of ensiling time on kernel fraction particle size distribution, geometric mean particle size, surface area and processing score in whole-plant corn silage harvested with a self-propelled forage harvester.

Item	Ensiling Time (Day)			SEM <sup>5</sup>	p-Value
	0 d	35 d	140 d		
Sieve <sup>1</sup> , mm					
6.7	21.7 <sup>a</sup>	16.1 <sup>b</sup>	14.6 <sup>b</sup>	0.78	<0.01
4.75	20.7	22.1	21.9	0.66	0.30
3.35	14.7	14.5	14.0	0.23	0.13
2.36	18.8	18.4	19.0	0.40	0.55
1.70	8.1 <sup>b</sup>	9.4 <sup>a</sup>	10.0 <sup>a</sup>	0.21	<0.01
1.18	4.8 <sup>b</sup>	6.3 <sup>a</sup>	6.6 <sup>a</sup>	0.24	<0.01
0.59	6.0 <sup>b</sup>	7.2 <sup>a</sup>	7.3 <sup>a</sup>	0.22	<0.01
Pan	5.0 <sup>b</sup>	5.9 <sup>a</sup>	6.6 <sup>a</sup>	0.21	<0.01
% < 4.75 <sup>2</sup>	57.6 <sup>b</sup>	61.8 <sup>a</sup>	63.5 <sup>a</sup>	1.01	<0.01
GMPS <sup>3</sup> , µm	3420.0 <sup>a</sup>	3059.5 <sup>b</sup>	2981.8 <sup>b</sup>	49.07	<0.01
Surface area, cm <sup>2</sup> g <sup>-1</sup>	17.5 <sup>b</sup>	19.3 <sup>a</sup>	19.7 <sup>a</sup>	0.27	<0.01
Processing score <sup>4</sup>	49.0 <sup>b</sup>	56.4 <sup>ab</sup>	58.9 <sup>a</sup>	2.37	0.02

<sup>1</sup> Percentage of particles retained on each sieve (DM basis). <sup>2</sup> Proportion of kernel fraction passing through a 4.75 mm sieve. <sup>3</sup> GMPS: geometric mean particle size. <sup>4</sup> Percent of starch passing through a 4.75 mm sieve, measured as described by Ferreira and Mertens [23]. <sup>5</sup> SEM: standard error of mean. <sup>a,b</sup> Means in the same row with different superscript letters differ by Tukey test ( $p \leq 0.05$ ).

There was a trend for interaction between the TLOC and ensiling time for the kernels in the pan ( $p = 0.10$ ; Figure 2). In the fresh samples and 140 d of ensiling, the TLOC of 6 mm promoted the greatest amount retained in the pan; however, at 35 d, no differences occurred among the treatments. A higher percentage of kernels smaller than 4.75 mm and with less surface area was observed for the 35 and 140 d ensiling in comparison with the fresh samples. The GMPS was reduced in both ensiling times. The CSPS was different only between the fresh (0 d) and 140 d of ensiling (49.0 vs. 58.9%). There were no more interactions ( $p > 0.12$ ; interaction's  $p$ -values were not shown in tables or figures).





**Figure 2.** Interaction between theoretical length of cut and ensiling time (day) for kernels retained in the pan ( $p = 0.10$ ; SEM = 0.21) in whole-plant corn silage harvested with a self-propelled forage harvester. SEM: standard error of mean. <sup>a,b</sup> Means in the same ensiling time with different superscript letters differ by Tukey test ( $p \leq 0.05$ ).

#### 4. Discussion

The nutrient composition of whole-plant corn (% DM)—30.6% starch and 43.6% NDF—represents high-quality corn silage by international standards [26]. The lack of differences in the particle size distribution and MPL between the TLOCs of 3 and 6 mm was not expected in the PTFH. The TLOC is controlled by the peripheral speed of the feed rolls relative to the speed of the cutter head and the number of cutter head knives [16]. The number of knives inside the PTFH and the speed of the cutter head were the same in both of these treatments. The only way to change the TLOC is by altering the peripheral speed of the feed rolls, relative to the cutter head speed, by changing the drive sprockets or gears driving the feed rolls [16].

The company's gear setting recommendations to reach different particle sizes at TLOCs of 3 and 6 mm did not achieve this objective. However, the TLOC of 9 mm promoted an increase in particles retained on the top two PSPS sieves, and, as a consequence, the MPL increased. Shinnars et al. [9] evaluated the effect of three TLOC settings (12.7, 19.0 and 25.4 mm) on WPCS particle size in a PTFH with a KP at a roll gap of 1 mm. In this trial, the particle size was determined by an apparatus and procedure described in the American Society of Agricultural Engineers—ASAE standard S424.1 [27]. A higher percentage of particles retained in an 18 mm sieve was observed followed by an increase in the MPL from 11.0 to 13.4 mm.

In the second experiment, we observed that the increase in the TLOC from 6 to 18 mm led to a greater percentage of longer particles (>19 mm) and MPL. Johnson et al. [28] also reported increments of MPL and the percentage of longer particles in response to different TLOC settings (11.1, 27.8 and 39.7 mm) for WPCS harvested by SPFH. Ferraretto et al. [13] studied the impact of three hybrids at two maturities (milk-line and 7 d later) and at two TLOC settings (6.4 mm or 19.5 mm) on the particle size and processing score. The particle size was increased with longer TLOC settings, as expected. The samples harvested with a 6.4 mm TLOC, on average, showed less retention in the coarse (3.5 vs. 9.0%) and medium (52.9 vs. 58.1%) sieves, but more material retained in the fine sieve (41.3 vs. 30.6%) than the 19.5 mm TLOC samples. The material in the bottom pan did not differ between the TLOC settings and averaged 2.3%. The kernel processing score varied among all the samples, but it was not affected by the TLOC setting.

The ensiling time affected the WPCS particle distribution only in the second experiment. The longest ensiling time had a higher percentage of particles retained on the top two PSPS sieves; therefore, the MPL increased. At day 35 of ensiling, there was an increase of particles only in the 8 mm PSPS sieve. Johnson et al. [28] observed an increase in the MPL

and particles retained in the 19 mm sieve with an ensiling period. These authors suggested that compaction of the forage in the silo may have altered the physical characteristics of the forage (flattened some of the particles), making it difficult for some particles to pass through the screens in the PSPS. The reason for the interaction between the TLOC and ensiling time for particles smaller than 1.18 mm is unknown.

Reducing the TLOC can be a possible strategy for improving kernel breakage in PTFH without a KP [8]. Dias Junior et al. [29] reported a reduction in visible intact kernels in WPCS harvested by PTFH when the TLOC was reduced from 8.5 mm to 3 mm. In the present study, we expected a reduction in the kernel particle size for the smallest TLOC; however, this did not happen in the first experiment. The values of the CSPS were below 50% for the PTFH and from 51.1 to 60.4% for the SPFH samples. In this way, they can be classified as inadequately and adequately processed, respectively [30]. The hybrid tested had a high percentage of vitreous endosperm (65%), which is related to kernel hardness [6] and can make kernels more difficult to break by the harvester processors [2]. It could be worse for a PTFH without a KP, which depends on the cutting knives for kernel breakage. Moreover, kernel vitreousness traditionally increases throughout the maturity of WPCS [31], which may explain the decrease in TTSD for cows fed with very dry WPCS (>40% DM), even when processed [4]. This effect may occur in Brazilian vitreous corn hybrids at even lower than 40% DM.

The common TLOC used for SPFH is 19 mm in the USA [16,17]. However, even within a range of TLOC below this target (e.g., 12 and 18 mm), the CSPS was not good. Through a meta-analysis of 24 studies, Ferraretto and Shaver et al. [4] evaluated the impact of the TLOC and kernel processing on starch digestibility. The authors reported decreased ruminal starch digestibility when WPCS was harvested with a long TLOC (>32 mm) and proposed that the long fiber fraction could reduce kernel breakage during processing. This effect might be occurring at TLOC settings below 32 mm for vitreous corn hybrids. Research should be focused on evaluating strategies (i.e., new KP, roll speed differential, roll gap and TLOC settings) within a wide range of maturities to optimize kernel processing in vitreous endosperm hybrids.

The ensiling lengths of either 35 or 140 d reduced the GMPS even in poorly processed silage (CSPS < 26.4%) harvested by the PTFH. As a consequence, the surface area and proportion of kernels smaller than 4.75 mm increased. The same effect was observed for the SPFH samples with adequate CSPS. This impact of the ensiling time for these kernel fraction variables (GMPS and surface area) of WPCS has not been reported in the literature before. Furthermore, Dias Junior et al. [3] observed a strong correlation between these kernel variables and ruminal DM disappearance in unfermented kernels at different times of incubation (3, 6, 9 and 12 h); thus, changing the ensiling time to improve DM digestibility through reduction of kernel particle size is possible. Ferraretto et al. [14] reported, across two experiments, an increase of 7 to 10% for the CSPS in WPCS ensiled for at least 30 d and up to 240 d. In the present trial, the CSPS was not altered by the ensiling time in the PTFH samples but was increased by 9.9% after 140 d of storage in comparison with unfermented samples. These findings suggest that ensiling time of at least 140 d may be a good strategy to increase kernel damage.

Hoffman et al. [12] reported physical–chemical alterations in high-moisture corn silage due to a longer ensiling time (240 d). A reduction in the  $\alpha$ ,  $\gamma$ ,  $\delta$  and  $\beta$  prolamin-zein subunits of the starch-protein matrix was observed, followed by an increase in buffer-soluble CP at 240 d of ensiling in comparison with unfermented samples. Through electron micrograph pictures, the authors demonstrated that starch-zein protein clusters within particles of high-moisture corn were dissociated. This effect of proteolysis may be a possible explanation for the alterations in the GMPS, surface area, proportion of kernels smaller than 4.75 mm and CSPS.

## 5. Conclusions

In the PTFH trial, the TLOCs of 3 and 6 mm resulted in no differences in the WPCS particle size distribution. However, the TLOC of 9 mm increased the MPL. The increase in the TLOC in the SPFH led to a higher MPL and percentage of long particles (>19 mm). The ensiling period increased the MPL and longer particles only for the WPCS harvested by the SPFH. The strategy of reducing the TLOC in the SPFH was efficient to improve the percentage of kernels smaller than 4.75 mm. Furthermore, the TLOC of 6 mm led to the best kernel processing in the SPFH. The ensiling time reduced the particle size of the kernel fractions for both forage harvesters. The CPCS increased only with 140 d of ensiling and when the WPCS was harvested with the SPFH. These findings suggest that increasing ensiling time and a low TLOC in SPFH (6 mm) may be good strategies to increase kernel damage in Brazilian vitreous corn hybrid silages.

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