Relationship between processing score and kernel-fraction particle size in whole-plant corn silage

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ABSTRACT

Kernel processing increases starch digestibility in whole-plant corn silage (WPCS). Corn silage processing score (CSPS), the percentage of starch passing through a 4.75-mm sieve, is widely used to assess degree of kernel breakage in WPCS. However, the geometric mean particle size (GMPS) of the kernel-fraction that passes through the 4.75-mm sieve has not been well described. Therefore, the objectives of this study were (1) to evaluate particle size distribution and digestibility of kernels cut in varied particle sizes; (2) to propose a method to measure GMPS in WPCS kernels; and (3) to evaluate the relationship between CSPS and GMPS of the kernel fraction in WPCS. Composite samples of unfermented, dried kernels from 110 corn hybrids commonly used for silage production were kept whole (WH) or manually cut in 2, 4, 8, 16, 32 or 64 pieces (2P, 4P, 8P, 16P, 32P, and 64P, respectively). Dry sieving to determine GMPS, surface area, and particle size distribution using 9 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 pan, as well as ruminal in situ dry matter (DM) digestibilities were performed for each kernel particle number treatment. Incubation times were 0, 3, 6, 12, and 24 h. The ruminal in situ DM disappearance of unfermented kernels increased with the reduction in particle size of corn kernels. Kernels kept whole had the lowest ruminal DM disappearance for all time points with maximum DM disappearance of 6.9% at 24 h and the greatest disappearance was observed for 64P, followed by 32P and 16P. Samples of WPCS (n = 80) from 3 studies representing varied theoretical length of cut settings and processor types and settings were also evaluated. Each WPCS sample was divided in 2 and then dried at 60°C for 48 h. The CSPS was determined in duplicate on 1 of the split samples, whereas on the other split sample the kernel and stover fractions were separated using a hydrodynamic separation procedure. After separation, the kernel fraction was redried at 60°C for 48 h in a forced-air oven and dry sieved to determine GMPS and surface area. Linear relationships between CSPS from WPCS (n = 80) and kernel fraction GMPS, surface area, and proportion passing through the 4.75-mm screen were poor. Strong quadratic relationships between proportion of kernel fraction passing through the 4.75-mm screen and kernel fraction GMPS and surface area were observed. These findings suggest that hydrodynamic separation and dry sieving of the kernel fraction may provide a better assessment of kernel breakage in WPCS than CSPS.
Key words: corn silage, particle size, corn silage processing score

INTRODUCTION

Whole-plant corn silage (**WPCS**) is a high-energy forage with high DM yield per hectare from kernel and stover fractions, which are high in starch and NDF contents, respectively. The kernel fraction, composed of approximately 72% starch (DM basis; Huntington, 1997), represents nearly 45% of the whole-plant DM (Philippeau and Michalet-Doreau, 1998) and more than 50% of the energy in WPCS (calculated using NRC, 2001). Total-tract starch digestibility of WPCS, however, ranges from about 80 to 98% in lactating dairy cows fed WPCS-based diets (Ferraretto and Shaver, 2012b). Processing WPCS through rollers during harvest reduces the particle size of corn kernels and has been reported to increase total-tract starch digestibility (Bal et al., 2000; Johnson et al., 2002; Cooke and Bernard, 2005). Nevertheless, the efficacy of kernel processing in WPCS is variable and can be influenced by several factors, including maturity or DM content of WPCS at harvest, theoretical length of cut, type of processor, and processor roll-gap clearance (Shinners et al., 2000; Ferraretto and Shaver, 2012a,b). Therefore, methods to evaluate the adequacy of kernel processing in WPCS are critical.

Ferreira and Mertens (2005) established a methodology to determine degree of kernel breakage in WPCS defined as corn silage processing score (**CSPS**). This methodology is widely used in the dairy industry and measures the proportion of starch passing through a 4.75-mm sieve after vertical shaking separation procedures using dried WPCS samples. However, the use of a single sieve to determine degree of kernel processing leads to the assumption that all particles passing through this sieve are equal. It is unknown, however, if the proportion of different fragments comprising this pool of particles vary widely. Wide variation would suggest that differences in starch digestibility among WPCS samples of similar CSPS exist.

Mean particle size is a mathematical function that describes the degree of reduction of a set of particles and is defined by measuring the relative amounts of particles retained in varied sieves of known dimensions (Liu, 2008). Particle size distribution analysis is desirable in several fields that handle granular or particulate materials. Granulometry is a common method to determine the particle size distribution of feed materials (Clementson and Ileleji, 2012), which can be performed with dry sieving. The material passes through a series of sieves, which are individually weighed and the proportion of material retained on each sieve determined. Geometric mean particle size (**GMPS**) is the most common method used for comparison of particle size distribution in cereal grains (ASABE, 2007). Recently, GMPS has been suggested, in combination with zein protein or ammonia-N, for modeling ruminal and total-tract starch digestibilities and rate of ruminal starch digestion for dry and high-moisture corns (Hoffman et al., 2012a). The GMPS of the kernel fraction in WPCS has not been well described and may provide valuable insights for modeling starch digestibility in WPCS. Thus, the objectives of our study were to evaluate (1) particle size distribution and ruminal in situ DM disappearance of unfermented kernels cut in varied particle sizes; (2) a method to measure GMPS in WPCS kernels; and (3) the relationship between CSPS and hydrodynamically separated kernel fraction GMPS in WPCS.

MATERIALS AND METHODS

Unfermented Kernels

Unfermented kernels from 110 corn hybrids from varied genetic lines commonly used for silage production were obtained from previous experiments in our laboratory. Kernels were collected from middle portions of ears at the maturity stage between one-half and two-thirds of the milk line. All kernels were previously dried

at 40°C for 72 h in a forced-air oven. Kernels were kept whole (**WH**) or manually cut in 2, 4, 8, 16, 32, or 64 pieces (**2P**, **4P**, **8P**, **16P**, **32P**, and **64P**, respectively) using scalpel blades (Surgical blade stainless steel n.21, Feather Safety Razor Co., Ltd., Ozaka, Japan). The first cut was done across the kernel, and each subsequent cut halved a previously cut piece. A total of 330 kernels (3 per hybrid) were obtained for each particle number treatment. Throughout the manuscript these treatments will be referred to as a kernel particle number treatment. Each sample (128.2 $\pm$ 7.0 g; mean $\pm$ SD) was dry-sieved using a Tyler Ro-Tap Shaker (model RX-29, W.S. Tyler, Mentor, OH) using a set of 9 sieves (W.S. Tyler) with nominal square apertures of 9.50, 6.70, 4.75, 3.35, 2.36, 1.70, 1.18, and 0.59 mm and pan (ASABE, 2007) to determine particle size distribution; GMPS (μm) and surface area (cm^2/g) were calculated using a log normal distribution (Baker and Herrman, 2002). Sieves were a combination of sieves used for CSPS (Ferreira and Mertens, 2005; 9.50-, 6.70-, 4.75-, 2.36-, 1.18-, and 0.59-mm sieves) or dry ground corn GMPS (Baker and Herrman, 2002; 3.35- and 1.70-mm sieves). The associated use of these 2 sets of sieves was chosen in an attempt to better characterize and describe the particle size distributions (% of DM retained on each sieve).

Subsequently, in situ measurements of ruminal DM disappearance were performed for each kernel particle number treatment using 2 ruminally cannulated, mid-lactation, multiparous Holstein cows fed a TMR containing (DM basis) alfalfa silage (44.5%), nonexperimental corn silage (26.8%), alfalfa hay (10.7%), wheat straw (6.5%), and concentrate mixture (11.5%). Individual samples of each kernel particle number treatment were recombined from each of the sieves after dry sieving procedures and evaluated for DM disappearance. Dacron polyester cloth bags (R510, 10 × 20 cm and 50-μm pores; Ankom Techonology, Macedon, NY) containing approximately 5 g of DM of each unfermented kernel particle number treatment, yielding a ratio of sample mass per bag area of 25 mg/cm^2, were incubated in duplicate within each cow. The in situ bags for the respective treatments for each time point (3, 6, 12, and 24 h) were placed in a nylon laundry bag (30 × 40 cm) and then positioned in the ventral rumen of each cow. Each laundry bag was attached to the inside of the rumen cannula with a 75-cm long nylon rope and contained a rubber weight to ensure they remained submerged in the ruminal contents. Bags were moistened in warm water for 30 s before incubation. Each laundry bag contained a blank bag to allow correction for infiltration of DM into sample bags. After removal, samples were soaked in cold water for 15 min before washing twice in

a commercial washing machine (Whirlpool #3347019A, Whirlpool Corp., Benton Harbor, MI) using the regular cycle rinse setting with cold water for 12-min cycles (Cherney et al., 1990). Two bags for each treatment (0-h bags) were soaked for 30 min in tepid water and washed with the rest of the sample bags. Bags were dried together in a forced-air oven at 60°C for 48 h. The mean DM disappearance value of the 2 bags on a treatment per incubation time was calculated for each cow.

In situ data were analyzed using PROC MIXED (SAS Institute, 2004). The model included the fixed effect of treatment, hour, and the interaction of treatment and hour, and cow as a random effect. Statistical analyses were performed on 0-h bags using PROC MIXED (SAS Institute, 2004) and the fixed effect of treatment. Means were determined using the least squares means statement and treatment means were compared using the Bonferroni's t-test option after a significant overall treatment F-test. Bonferroni's t-test is a sequentially rejective test based on the Holm-Bonferroni method (Holm, 1979). Interaction effects were partitioned using the SLICE option (SAS Institute, 2004). Subsequently, regression analysis was performed using PROC REG of SAS (SAS Institute, 2004) to determine linear and quadratic relationships between ruminal in situ DM disappearance and GMPS or surface area. Analyses were performed on all the individual time point treatment means. Best fit regression (linear or quadratic) was chosen using the highest coefficient of determination (R^2) and lowest root mean square error (**RMSE**) as indicators. Statistical significance and tendencies were declared at $P \leq 0.05$ and $P > 0.05$ to $P < 0.10$, respectively.

WPCS

Eighty WPCS samples from 2 feeding trials (Ferraretto and Shaver, 2012a; Vanderwerff et al., 2015) and a field survey of commercial farms (Salvati et al., 2015) were selected to represent varied corn silage harvesting equipment, processor types, and settings. Samples were frozen and stored at −20°C in all 3 studies and thawed overnight before sample analysis. Each WPCS sample was placed in a bucket, homogenized manually, and divided into two 1-kg subsamples using a quartering technique: homogeneous samples were divided into 4 equal subsamples. Two subsamples allocated diagonally were rehomogenized and saved as 1 subsample, whereas the other 2 subsamples were rehomogenized and saved as the other subsample. Each subsample was dried at 60°C for 48 h in a forced-air oven. On 1 of the subsamples, CSPS was determined in duplicate as described by Ferreira and Mertens (2005). On the

other subsample, kernel and stover fractions were separated by a hydrodynamic separation procedure (Savoie et al., 2004). This method is based on differences in buoyancy between the kernels and stover. 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 such that the entire sample was submerged in water. After 2 min, the stover fraction, which floats due to a lower density than water, was removed gently using a small 1.17-mm sieve (Savoie et al., 2004). Subsequently, the water was poured out of the tub, retaining the kernel fraction, which has a density greater than water (Savoie et al., 2004). After separation, the kernel fraction was transferred to aluminum plates, redried at 60°C for 48 h in a forced-air oven, and dry sieved using a Tyler Ro-Tap Shaker (model RX-29, W.S. Tyler) with a set of 9 sieves having nominal square apertures of 9.50, 6.70, 4.75, 3.35, 2.36, 1.70, 1.18, and 0.59 mm and pan; GMPS (μm) and surface area (cm^2/g) were calculated using a log normal distribution (Baker and Herrman, 2002). All samples were recombined and ground to pass through a 1-mm screen of a Wiley mill (Thomas Scientific, Swedesboro, NJ) for starch, NDF, and CP determination on hydrodynamically separated WPCS kernel fraction by near-infrared reflectance spectroscopy (**NIRS**; model 6500; Foss-NIR System, Silver Spring, MD). Each sample was packed into a cylindrical sample holder equipped with a quartz window (Part number 60013287; Foss-NIR System) and scanned, between 400 and 2,498 nm, as described by Mentink et al. (2006). Prediction equations for determining nutrient composition were obtained from the NIRS Forage and Feed Test Consortium website (http://nirsconsortium.org/). These equations were calibrated for CP (methods 984.13, 988.05, and 990.03; AOAC International, 2012), NDF assayed with α-amylase and sodium sulfite (method 2002.04; AOAC International, 2012) and expressed inclusive of residual ash (**aNDF**), and starch (Bach Knudsen, 1997) using 754 ($R^2 = 0.88$), 1,193 ($R^2 = 0.93$), and 320 ($R^2 = 0.97$) fermented WPCS samples, respectively, from participating laboratories from all over the United States and collected over more than 20 yr.

Regressions to determine linear and quadratic relationships between CSPS in WPCS and hydrodynamically separated WPCS kernel fraction GMPS, surface area, and retained proportion on sieves of varied particle size were performed using PROC REG of SAS (SAS Institute, 2004). Best fit regression (linear or quadratic) was chosen using the highest R^2 and lowest RMSE as indicators. Statistical significance and tendencies were declared at $P \leq 0.05$ and $P > 0.05$ to $P \leq 0.10$, respectively.

DIAS JUNIOR ET AL.

Table 1. Particle size distribution, geometric mean particle size (GMPS), and surface area of unfermented kernels[1]

Item	WH	2P	4P	8P	16P	32P	64P
Sieve,[2] mm							
9.50	0.00	0.00	0.00	0.00	0.00	0.00	0.00
6.70	79.71	20.71	0.63	0.00	0.00	0.00	0.00
4.75	20.29	78.16	60.50	5.57	0.14	0.00	0.00
3.35	0.00	0.67	38.34	87.98	15.81	0.57	0.00
2.36	0.00	0.00	0.20	5.01	74.23	53.74	3.97
1.70	0.00	0.00	0.02	0.18	7.78	43.42	65.01
1.18	0.00	0.07	0.04	0.06	0.69	1.68	20.35
0.59	0.00	0.18	0.10	0.67	0.90	0.28	6.80
Pan	0.00	0.21	0.15	0.52	0.45	0.31	3.85
GMPS, μm	7,556.7	6,074.9	4,980.8	3,953.4	2,866.7	2,413.7	1,683.3
Surface area, cm^2/g	3.6	6.5	8.9	11.9	15.9	17.8	23.2

[1]Kernels were kept whole (WH) or broken in 2, 4, 8, 16, 32, or 64 pieces (2P, 4P, 8P, 16P, 32P, and 64P, respectively).

[2]Percentage of particles retained on each sieve (DM basis).

Simulations were performed using the Feed Grain V2.0 Evaluation System (Hoffman et al., 2012b) to predict the potential effect of GMPS on extents of ruminal and total-tract starch digestibilities and ruminal rate of starch digestibility for dairy cows. The full model description can be accessed in Hoffman et al. (2012c). Briefly, the Feed Grain V2.0 Evaluation System is based on the model proposed by Hoffman et al. (2012a) and uses GMPS and ammonia-N or prolamin to predict starch digestibility of HMC or dry corn, respectively. Hydrodynamically separated WPCS kernel fraction GMPS measurements from all samples were input in the model at a constant ammonia-N concentration value (3.3 ± 2.2% CP). Ammonia-N concentration was arbitrarily chosen based on the average of ammonia-N values of high-moisture corn samples with a similar storage length (Ferraretto et al., 2014).

RESULTS AND DISCUSSION

Unfermented Kernels

Particle size distribution of unfermented kernels is in Table 1. As designed, the distribution of particles among the different sieves varied widely among kernel particle number treatments. It was observed that each sieve retained more than one kernel particle number treatment. The least numeric variation was observed for 8P with 88% of the particles retained on the 3.35-mm sieve. In contrast, 64P was the most numerically variable with material retained on 2.36-, 1.70-, 1.18-, and 0.59-mm sieves, and pan. This is related to wide variation in kernel size comprising the pool of hybrids evaluated and the corresponding difference in particle size when small and large kernels were cut in 64P. Kernel size naturally varies within and across hybrids (Graven and Carter, 1990) and may be affected by vary-

ing vigor, environmental conditions during pollination and fill period, kernel position in the ear, and parent tendencies (EnayatGholizadeh et al., 2012; Mondo et al., 2013). In addition, kernels were cut by hand; therefore, samples from the same kernel may vary in particle size. Ferreira and Mertens (2005) proposed CSPS, the proportion of starch passing through a 4.75-mm sieve, as a quantitative measure of kernel fragmentation in WPCS. Those authors suggested that CSPS represents the proportion of kernels that are broken in at least one-fourth of a kernel. Our results, however, revealed that 60% of 4P kernels were retained on the 4.75-mm sieve. Perhaps the use of a broader hybrid pool and use of dried, unfermented kernels in the present study may explain some of the differences between studies.

Ruminal in situ DM disappearance of each particle number treatment of unfermented kernels is in Table 2. Digestibility of DM in corn kernels is strongly related to starch digestibility (Philippeau and Michalet-Doreau, 1999; Ngonyamo-Majee et al., 2009), and thus served as a proxy for starch digestibility in the present study. Treatments followed a similar pattern across all time points, but a treatment × time interaction was observed ($P = 0.01$), as the magnitude of response over time was different for the different treatments. A negative quadratic relationship was observed between GMPS and ruminal in situ DM disappearance for all time points (Figure 1; $P < 0.01$), with the lowest R^2 observed at 3 h ($R^2 = 0.89$). Overall, ruminal DM disappearance increased ($P < 0.01$) with the reduction in particle size of corn kernels. Likewise, a positive quadratic relationship was observed between surface area and ruminal in situ DM disappearance for all time points (Figure 2; $P < 0.01$), with lowest R^2 observed at 3 h of incubation ($R^2 = 0.89$). Reduced GMPS increased total-tract starch digestibility in dry ground and high-moisture corn (Ferraretto et al., 2013). Kernels kept whole had the lowest
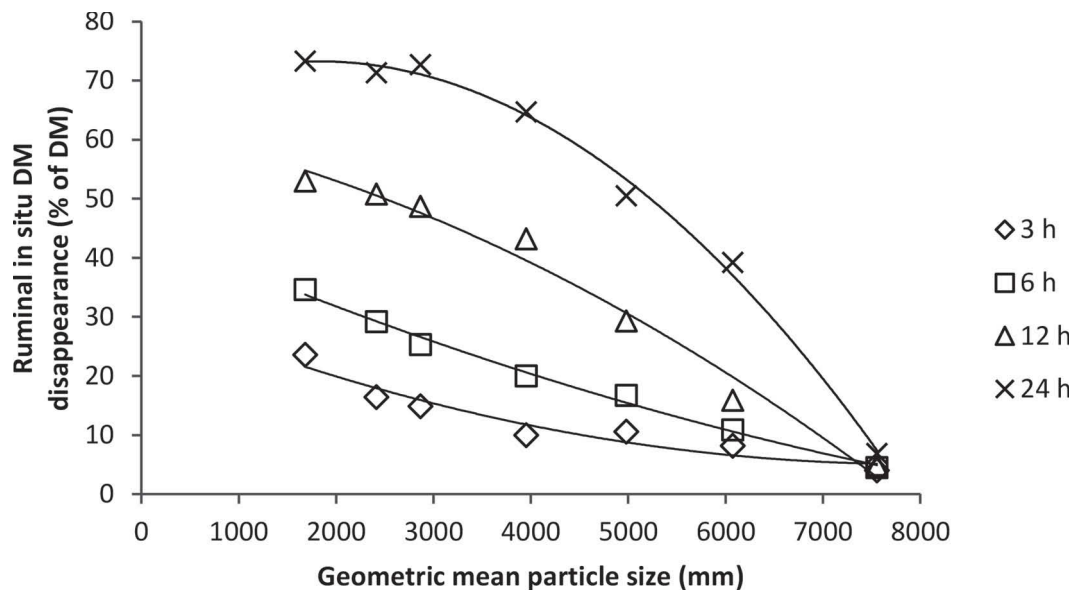
Figure 1. Relationship between ruminal in situ DM disappearance (% of DM) and geometric mean particle size (μm) in unfermented kernels. Prediction equation for 3 h: $y = 31.58\ (\pm4.53) - 0.007x\ (\pm0.01) - 4.170x^2\ (\pm2.42)$; n = 7, root mean square error (RMSE) = 2.10, R^2 = 0.89, P = 0.01. Prediction equation for 6 h: $y = 45.11\ (\pm2.42) - 0.007x\ (\pm0.01) - 2.428x^2\ (\pm1.21)$; n = 7, RMSE = 1.05, R^2 = 0.99, P = 0.001. Prediction equation for 12 h: $y = 62.13\ (\pm6.74) - 0.004x\ (\pm0.01) - 5.909x^2\ (\pm3.60)$; n = 7, RMSE = 3.12, R^2 = 0.97, P = 0.001. Prediction equation for 24 h: $y = 66.65\ (\pm4.47) + 0.001x\ (\pm0.01) - 0.001x^2\ (\pm2.39)$; n = 7, RMSE = 2.07, R^2 = 0.99, P = 0.001.

ruminal DM disappearance for all time points, with a maximum DM disappearance of 6.9% at 24 h. The pericarp layer surrounding the starch endosperm is highly resistant to microbial attachment (McAllister et al., 1994) and likely inhibited DM disappearance in WH. The greatest ruminal in situ DM disappearance was observed for 64P, followed by 32P and 16P, followed by a steady decrease in DM disappearance in 8P, 4P, 2P, and WH. As kernel fragmentation increases, greater surface area is available for attachment of rumen microorganisms to starch granules and enzymatic digestion (McAllister et al., 1993). Furthermore, reduced GMPS attenuates the negative effects of the protein matrix encapsulating starch granules (Hoffman et al., 2011), thereby resulting in greater starch digestion. Currently,

the most common method used to assess degree of kernel processing in WPCS is CSPS, which considers all particles passing through a 4.75-mm sieve to be equal in digestibility characteristics. Nevertheless, dry sieving revealed that 4P, 8P, 16P, 32P, and 64P unfermented kernel fractions with different in situ disappearance passed through this sieve. Differences in DM disappearance observed in the present study emphasize the importance of a more descriptive particle size methodology for kernels. An important aspect to consider is the use of unfermented kernels in the present study, which are well known to be less digestible than fermented kernels (Philippeau and Michalet-Doreau, 1998; Hoffman et al., 2011). Perhaps differences in DM disappearance among the varied unfermented kernel particle number treat-

Table 2. Ruminal in situ DM disappearance (% of DM) of unfermented kernels[1,2,3]

Time point, h	WH	2P	4P	8P	16P	32P	64P	SEM[4]	P-value
0	4.6[d]	4.8[d]	6.1[c]	4.9[d]	6.8[bc]	7.5[b]	10.0[a]	0.5	0.01
3	4.0[c]	8.2[b]	10.6[b]	10.0[b]	14.9[b]	16.4[ab]	23.6[a]	6.1	0.03
6	4.5[d]	10.9[c]	16.7[bc]	20.0[b]	25.3[ab]	29.2[a]	34.6[a]	6.1	0.001
12	4.9[f]	15.9[e]	29.3[de]	43.2[d]	48.7[c]	50.8[b]	53.0[a]	5.3	0.001
24	6.9[e]	39.2[d]	50.5[c]	64.7[b]	72.7[ab]	71.3[a]	73.3[a]	6.1	0.001

[a–e]Means in the same row with different superscripts differ ($P \leq 0.05$).

[1]Kernels were kept whole (WH) or broken in 2, 4, 8, 16, 32, or 64 pieces (2P, 4P, 8P, 16P, 32P, and 64P, respectively).

[2]Hour ($P = 0.001$) and treatment by hour ($P = 0.001$) effects.

[3]Blank bag weights before and after incubations were 3.07 (±0.08) and 3.10 g (±0.08), respectively.
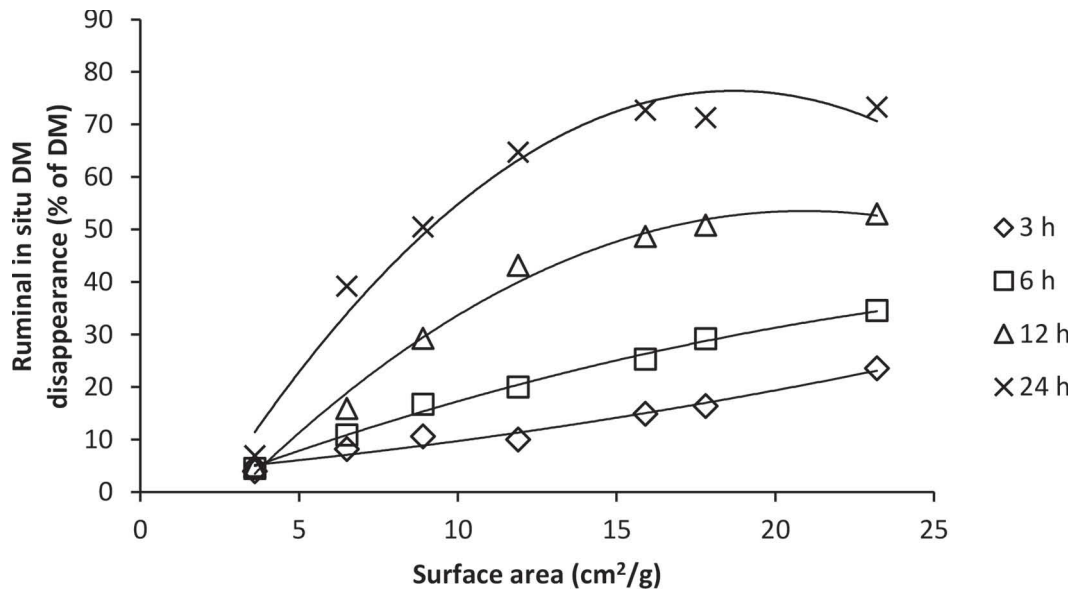
[4]Greatest standard error of the mean.

Figure 2. Relationship between ruminal in situ DM disappearance (% of DM) and surface area (cm^2/g) in unfermented kernels. Prediction equation for 3 h: $y = 3.15 (\pm2.25) + 0.507x (\pm0.39) + 0.015x^2 (\pm0.01)$; n = 7, root mean square error (RMSE) = 0.40, R^2 = 0.95, P = 0.001. Prediction equation for 6 h: $y = -3.186 (\pm1.50) + 5.130x (\pm0.38) - 0.117x^2 (\pm0.01)$; n = 7, RMSE = 0.93, R^2 = 0.99, P = 0.001. Prediction equation for 12 h: $y = -19.44 (\pm3.87) + 6.979x (\pm0.66) - 0.167x^2 (\pm0.02)$; n = 7, RMSE = 2.41, R^2 = 0.98, P = 0.001. Prediction equation for 24 h: $y = -23.30 (\pm7.40) + 10.660x (\pm1.27) - 0.285x^2 (\pm0.05)$; n = 7, RMSE = 4.60, R^2 = 0.96, P = 0.001.

ments would be lower after ensiling. However, negative effects of increased particle size on starch digestibility by dairy cows was reported for both dry and ensiled corn in a recent meta-analysis (Ferraretto et al., 2013).

WPCS

Descriptive statistics for parameters measured on WPCS and hydrodynamically separated WPCS kernel fraction samples are in Table 3. As expected based on the diversity of the sample set, all parameters varied widely. The DM content of WPCS averaged 34.3% ranging from 25.5 to 47.1%, whereas starch content averaged 33.6% with a range of 17.1 to 42.6%. The large variance in DM and starch contents among WPCS samples was reflective of various environmental factors and harvesting practices from the source trials and farms. The CSPS of WPCS samples averaged 66.2% and ranged from 49.5 to 82.6%. Zwald et al. (2008), in a study with 81 WPCS samples from varied hybrids and planting and harvest dates, reported an average CSPS of 58.9% with a range of 24.5 to 89.7%. Several factors, including endosperm properties (Correa et al., 2002), maturity (Johnson et al., 2002), and degree of processing (Bal et al., 2000) of WPCS alter the fragmentation of kernels and thereby the digestibility of starch (Cooke and Bernard, 2005). The CSPS values observed in the present study reflect the large variation that exists in

the degree of kernel processing in WPCS. Starch content of the hydrodynamically separated WPCS kernel fraction averaged 70.6% with an 11.8-percentage-unit range (DM basis), giving a composition similar to those reported in other studies for the kernel fraction. These data highlight that the hydrodynamically separated WPCS kernels did not have contamination from other fractions. Ferreira and Mertens (2005) reported an average starch content of 63.7% (DM basis) in WPCS kernels that were hand-selected during the development of the CSPS procedure. Philippeau and Michalet-Doreau (1997) and Ngonyamo-Majee et al. (2008), studying the chemical and physical characteristics of diverse corn hybrids, reported average starch concentrations of 65.0 and 70.8% (DM basis), respectively, in unfermented WPCS kernels. In the present study, concentrations of CP and NDF in the WPCS kernel-fraction were 5.6 ± 1.3 and $11.0 \pm 1.7\%$, respectively. Ferraretto et al. (2014) reported CP and NDF concentrations in high-moisture corn to be 8.1 ± 1.7 and $7.1 \pm 1.4\%$, respectively, in a retrospective study containing more than 6,000 samples. Further research is warranted to evaluate the percentage of WPCS represented by hydrodynamically separated WPCS kernel fraction.

Relationships between WPCS CSPS and hydrodynamically separated WPCS kernel fraction particle size measurements are in Table 4. Linear relationships between CSPS and hydrodynamically separated WPCS

kernel fraction GMPS ($R^2 = 0.11$; $P = 0.01$) or surface area ($R^2 = 0.06$; $P = 0.03$) were poor. A strong quadratic relationship ($R^2 = 0.96$) between CSPS and starch GMPS was previously reported by Zwald et al. (2008). A positive linear relationship between WPCS CSPS and the proportion of hydrodynamically separated WPCS kernel fraction passing through the 4.75-mm screen was observed in the present study although the relationship was weak ($R^2 = 0.34$; $P = 0.001$). A distribution plot of CSPS and proportion of hydrodynamically separated WPCS kernel fraction passing through the 4.75-mm sieve (Figure 3) suggests that CSPS may underestimate starch passage through the 4.75-mm sieve. Because the CSPS uses a dried whole sample, fine starch particles may be retained on coarse fiber particles. This may explain the strong quadratic relationship observed by Zwald et al. (2008) when measuring starch GMPS in whole samples. In the study of Zwald et al. (2008), NIRS was used to predict particle size of starch within undried, unground corn silage and verified that some physical characteristics also interfered in the accuracy of predictions. When spectra was obtained from samples without particles greater than 19 mm the prediction was improved. According to those authors, long forage particles and cobs retained on a 19-mm sieve could contain some fine particles of starch that might influence the capability of NIRS to properly predict the physical characteristics of the kernel-fraction of WPCS. Our data showed that the CSPS was inversely related to hydrodynamically separated WPCS kernel fraction material retained on 4.75-mm ($R^2 = 0.26$; $P = 0.001$) sieves, as expected. Evaluation of fractions retained on individual sieves below the 4.75-mm sieve revealed a positive, but poor relationships between CSPS and 3.35- ($R^2 = 0.20$; $P < 0.01$), 2.36- ($R^2 = 0.47$; $P < 0.01$), and 1.70-mm ($R^2 = 0.14$; $P < 0.01$) sieves and pan ($R^2 = 0.26$; $P = 0.02$). Corn silage processing score was not related ($P > 0.10$) to hydrodynamically separated WPCS kernels retained on 1.18- and 0.59-mm sieves.

Among all evaluated hydrodynamically separated kernel fractions in WPCS, the particles retained on the 2.36-mm sieve, not the 4.75-mm screen, had the highest relationship with CSPS. Thus, the proportion of particles retained on this screen was evaluated as a possible single screen predictor of GMPS and surface area of hydrodynamically separated WPCS kernels (Table 5). A quadratic relationship was observed ($P = 0.01$), but the relationship was relatively weak for both GMPS ($R^2 = 0.16$) and surface area ($R^2 = 0.06$). Zwald et al. (2008) also observed low prediction of CSPS when using data of WPCS particles retained on a single screen. In addition, those authors reported difficulties using NIRS to predict particles retained on 4.75- and 3.35-mm vertical shaker screens.

The relationship between the proportion of hydrodynamically separated WPCS kernel particles passing through the 4.75-mm screen and hydrodynamically separated GMPS of WPCS kernels or surface area are in Table 5. Strong quadratic relationships between pro-

Table 3. Descriptive statistics of nutrient composition and corn silage processing score (CSPS) in whole-plant corn silage and hydrodynamically separated kernel-fraction nutrient composition, particle size distribution, surface area, and geometric mean particle size (GMPS)

Item	Mean	SD	Minimum	Maximum
Whole-plant corn silage				
DM, % (as fed)	34.3	3.9	25.5	47.1
Starch, % of DM	33.6	5.6	17.1	42.6
CSPS,[1] % starch passing through a 4.75-mm sieve	66.3	6.1	49.5	82.7
Kernel fraction				
CP, % of DM	5.6	1.3	3.8	9.5
NDF, % of DM	11.0	1.7	7.7	16.6
Starch, % of DM	70.6	2.80	63.9	75.7
GMPS, μm	2,637.5	862.7	1,614.9	7556.7
Surface area, cm^2/g	21.4	4.2	3.6	28.4
Kernel fraction retained on each sieve, %[2]				
6.70	5.5	4.4	0.4	25.7
4.75	20.7	5.7	8.7	33.5
3.35	18.6	3.3	11.1	30.0
2.36	15.3	2.8	9.1	23.8
1.70	11.5	1.6	6.9	15.1
1.18	8.9	1.6	5.7	14.5
0.59	10.4	2.4	5.8	19.2
Pan	9.0	2.8	3.6	18.3

[1]CSPS as in Ferreira and Mertens (2005).

[2]None of the samples had material retained in the 9.50-mm sieve.

Table 4. Linear relationship between corn silage processing score (% starch passing through a 4.75-mm sieve) in whole-plant corn silage and hydrodynamically separated kernel-fraction geometric mean particle size (GMPS), surface area, and retained proportion on sieves of varied particle size

	Parameter estimates				Model statistics		
Predicted	Intercept	SE	Slope	SE	P-value	R^2	RMSE[1]
GMPS, μm	3,969.92	479.52	−22.61	3,969.92	0.01	0.11	389.84
Surface area, cm^2/g	15.70	2.94	0.10	15.70	0.02	0.06	2.38
Particles retained,[2] mm							
4.75	52.50	6.10	−0.482	52.50	<0.01	0.26	4.95
3.35	2.55	3.65	0.242	2.55	<0.01	0.20	2.97
2.36	−5.38	2.50	0.312	−5.38	<0.01	0.47	2.03
1.70	5.11	1.82	0.096	5.11	<0.01	0.14	1.48
1.18	6.35	1.92	0.039	6.35	0.18	0.02	1.55
0.589	5.77	2.95	0.070	5.77	0.12	0.03	2.39
Pan	0.82	3.40	0.123	0.82	0.02	0.07	2.75
<4.75	67.75	4.45	0.403	67.75	<0.01	0.34	3.61

[1]Root mean square error.

[2]Percentage of particles retained on each sieve (DM basis).

portion of hydrodynamically separated WPCS kernel fraction passing through the 4.75-mm screen and GMPS ($R^2 = 0.69$; $P < 0.01$) and surface area ($R^2 = 0.54$; $P < 0.01$) were observed. Baker and Herrman (2002) found that surface area of kernel particles is an exponential function of GMPS. In our study, an increase in particles passing through 4.75-mm sieve was strongly associated with a decrease in the GMPS. Nevertheless, the relationship between hydrodynamically separated WPCS kernel particles passing through the 4.75-mm sieve and

Figure 3. Relationship between corn silage processing score (CSPS; % of starch passing through a 4.75-mm sieve) and proportion of hydrodynamically separated kernel fraction passing through a 4.75-mm sieve in whole-plant corn silage. Corn silage processing score prediction equation: y = 37.55 (±4.54) + 0.39x (±0.06); n = 80, root mean square error = 4.96, $R^2 = 0.34$, $P < 0.01$.

surface area was lower. This observation was expected because samples containing different proportions of varied particle sizes result in different surface area values. The particle size of kernel fractions in WPCS is important because it can influence the digestion of starch, which is the main source of digestible energy in WPCS. High demand exists for improved accuracy of predicting WPCS starch digestibility using laboratory assays, such as CSPS. Considering the great diversity in shape and size of the kernels in hybrids used for WPCS production, to define kernel processing based on the proportion of starch that passes through the 4.75-mm sieve may not be accurate. However, CSPS remains a useful tool to evaluate kernel processing in WPCS until a new method is fully adopted by commercial laboratories. Additionally, the use of a single larger screen may lead to an underestimation of the kernel digestibility; whereas the use of a single smaller screen may overestimate the digestion potential of the kernel fraction of WPCS. Total-tract starch digestibility of corn silage-based diets is highly variable (Ferraretto and Shaver, 2012b). This variation can be partially explained by the particle size of the kernel fraction. However, building relationships between digestibility and degree of kernel fragmentation is challenging due to lack of quantitative particle size data.

Simulation results of starch fermentation rate as well as ruminal and total-tract starch digestibility are in Figure 4A, 4B, and 4C, respectively. The predicted starch fermentation rate ranged from 12.9 to 19.8%/h. The model predicted ruminal and total-tract starch digestibility had differences of 10.6 and 3.4 percentage units, respectively, between minimum and maximum reported values. The understanding of fermentation rate and starch digestibility of WPCS would allow dairy
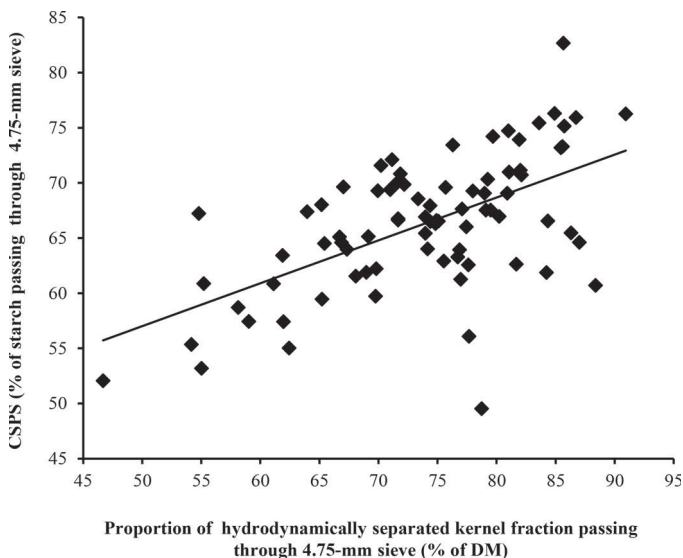
nutritionists to optimize diet formulation and thereby better meet the nutrient demands of high-producing dairy cows.

CONCLUSIONS

Particle size distribution of unfermented corn kernels cut in varied particle sizes revealed that only 40% of the kernels cut in 4 pieces passed through the 4.75-mm sieve. Measurements of ruminal in situ DM disappearance indicated that kernels of smaller particle
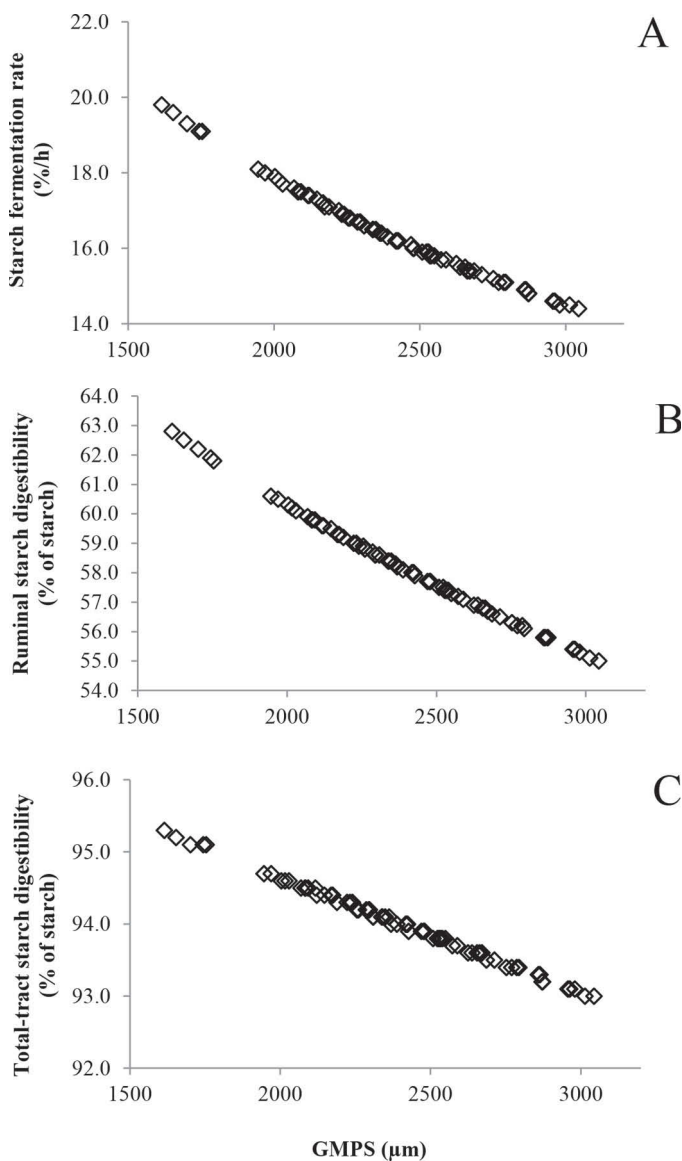

Figure 4. Simulations of the effect of kernel fraction geometric mean particle size (μm) on starch fermentation rate (%/h; A) and ruminal and total-tract starch digestibilities (% of starch; B and C, respectively) performed using the Feed Grain V2.0 Evaluation System (Hoffman et al., 2012b).

Table 5. Quadratic relationships between hydrodynamically separated kernel particles retained at the 2.36-mm sieve or passing through the 4.75-mm sieve and hydrodynamically separated kernel-fraction geometric mean particle size (GMPS, μm) and surface area (cm²/g)

Predicted (y)	Predictor (x)	Parameter estimates				Model statistics		
		Intercept	SE	Slope	SE	P-value	R²	RMSE[1]
% of particles retained at the 2.36-mm sieve	GMPS	5,453.13	929.02	−346.38	121.07	<0.01	0.16	377.41
	GMPS²			9.60	3.89			
% of particles retained at the 2.36-mm sieve	Surface area	8.65	5.86	1.66	0.76	0.04	0.06	2.38
	Surface area²			−0.05	0.03			
% of particles passing the 4.75-mm sieve	GMPS	8,262.72	1,201.04	−124.93	33.98	<0.01	0.69	228.81
	GMPS²			0.62	0.24			
% of particles passing the 4.75-mm sieve	Surface area	0.57	8.79	0.41	0.25	<0.01	0.54	1.67
	Surface area²			−0.001	0.002			

[1]Root mean square error.

sizes have increased in situ DM disappearance. Consequently, the utilization of a single sieve to assess the potential digestibility of kernels in WPCS may be too inaccurate for use in ruminant nutrition models to estimate the nutritive value of WPCS. Hydrodynamically separated WPCS kernel-fraction GMPS and surface area were poorly related to CSPS. These data suggest that hydrodynamic separation and dry-sieving of the kernel-fraction may provide a better assessment of kernel breakage in WPCS. The use of these procedures in combination with the Feed Grain V2.0 Evaluation System may enhance the modeling of starch digestibility in WPCS and further research is warranted.

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