Relationship Between Ruminal Starch Degradation and the Physical Characteristics of Corn Grain [1]

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ABSTRACT: The objectives of this study were to determine the range of variation in the rate and extent of in situ ruminal starch degradation of 14 corns differing in vitreousness and to predict ruminal starch degradability by physical characteristics of corn grains. This study was conducted with eight dent and six flint corns. Ruminal starch degradability was determined by an in situ technique on 3-mm ground grains. Physical characteristics of corn grain were measured: hardness by grinding energy and particle size distribution, apparent and true densities, and specific surface area. Ruminal DM and starch degradabilities averaged 50 and 55.1% and varied from 39.7 to 71.5% and from 40.6 to 77.6%, respectively. Ruminal starch degradability averaged 61.9 and 46.2% in dent and flint types, respectively. The proportion of coarse particles (61.9 vs 69.6% for dent and flint, respectively), the apparent density (1.29 vs 1.36 g/cm^3 for dent and flint, respectively), and the specific surface area (.13 vs .07 m^2/g for dent and flint, respectively) varied with the vitreousness. Ruminal starch degradability could be predicted accurately by vitreousness (r^2 = .89) or by the combination of apparent density and 1,000-grain weight (R^2 = .91), a measurement faster than the vitreousness determination.

Key Words: Maize, Texture, Rumen, Digestion, Density, Hardness

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Introduction

Research in starch utilization by ruminants has been reviewed recently (Nocek and Tamminga, 1991; Owens et al., 1997). Although starch in cereal grain is almost completely digested in the whole digestive tract, the rate and extent of ruminal fermentation vary widely with grain source and cereal processing (Owens et al., 1986; Theurer, 1986; Huntington, 1997). The site of starch digestion has implications for the nature and amount of nutrients delivered to the animal.

Another way to manipulate the rate of starch degradation is by selecting cultivars. Barley cultivar affects in vitro dry matter disappearance (Kemalyan et al., 1989) and ruminal starch degradation (G. R. Khorasani, personal communication). Sorghum grain variety (Streeter et al., 1990a) and hybrid (Streeter et al., 1990b) also altered the site and extent of starch digestion. In a comparison of in vitro ruminal starch disappearance rates of sorghum cultivars, Kotarski et al. (1992) reported a faster disappearance rate for cultivars with a floury endosperm compared with a grain with a horny endosperm. The texture of the grain seems to play a major role in ruminal starch degradation, as we showed in situ with corn grains (Philippeau and Michalet-Doreau, 1997).

The texture of corn hybrids growing in Europe is dent or semi-flint. The objective of this study was to determine the range of variation of ruminal starch degradation among dent and flint corn grains varying in vitreousness by the in situ technique proposed by Nocek and Tamminga (1991) and Sauvant et al. (1994) and to seek predictors of ruminal starch degradation from physical characteristics of the grain.

Materials and Methods

This study was performed with 14 experimental hybrids of corns (*Zea mays* L.) differing in the texture of the endosperm, dent (eight corns) or flint (six corns). The growing characteristics of corn cultivars are given in Table 1. The texture of the endosperm was characterized by the grain vitreousness, which

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Table 1. Description of the growing characteristics of corn grains

Corn	Texture	Origin	Year
1	Dent	Crémone, Italy	1992
2	Dent	Champaign, IL	1994
3	Dent	Champaign, IL	1995
5	Dent	Longué, France	1996
6	Dent	Longué, France	1996
7	Dent	Longué, France	1996
15	Dent	Marmande, France	1996
17	Dent	Tinlhat, France	1995
9	Flint	Longué, France	1996
11	Flint	Longué, France	1996
12	Flint	La Menitrée, France	1996
13	Flint	Longué, France	1996
16	Flint	Chappes, France	1995
18	Flint	Tinlhat, France	1995

was determined by a mass method previously described (Philippeau and Michalet-Doreau, 1997). The proportion of horny endosperm was determined by weighing the horny endosperm fraction and the degermed grain, and the vitreousness represents the massic proportion of the horny endosperm in the degermed grain. This procedure was performed with 10 grains for each cultivar. The 1,000-grain weight was determined from the weight of two randomly selected samples of 500 grains. The 1,000-grain weight was adjusted for 15.0% moisture content.

Physical Measurements. Grain hardness was determined as grinding energy and as particle size distribution. A preliminary step consisted of conditioning grains in a climatic room to approximately 9.4% moisture (from 8.3 to 10.1%). Seventy grams was ground in a laboratory hammer mill (Retsch, ZM1 type, F. Kurt Retsch GMBH & Co. KG, Haan, Germany) fitted with a 3-mm aperture screen. A milling speed setting of 1,500 rpm was used. The electric power consumed for grinding was recorded in duplicate and the grinding energy was taken as an index of hardness. The ground samples underwent sonic sieving. Two grams were placed in a Fritsch analysette 28 apparatus (Fritsch GMBH, Idar-Oberstein, Germany) equipped with a series of four sieves, with 90-, 125-, 250-, and 500-μm aperture sizes. Samples were lifted by a vertical oscillated airstream and blown through the sieve holes. The particles were therefore subjected to a high acceleration, resulting from the high number of oscillations (3,000/min) and the large amplitude of the airstream. Sieving time was 1.3 min at a 30% amplitude setting. The material retained on each sieve was weighed. The proportion of particles larger than 250 μm was taken as representative of coarse particles, and the proportion of particles that passed through the 125-μm sieve was considered as representative of fine particles. The proportion of particles with diameters between 250 and 125 μm gave the proportion of intermediate particles. Three replicates were completed for each cultivar.

A gas multipycnometer (Quantachrome Corporation, 5 Aerial Way, Syosset, NY) with helium was used to determine the volume of grain samples as described by Chang (1988). Apparent or true densities of grain were defined as the ratio of weight to the apparent or true volume of the grain sample. The apparent grain volume including void spaces inside the grain was determined from 70 g of whole grain conditioned as previously to 9.4% moisture. For the determination of the true grain volume, the sample was ground in a hammer mill through a 3-mm screen as previously described, and ground samples were oven-dried at 40°C for 48 h. The true volume was determined from 8 g of ground grain. Two replicates were done for each determination and each cultivar.

The specific surface area was determined by the BET gas adsorption principle (Brunauer et al., 1938). The method was based on the flowing-gas technique (Gemini 2360 analyzer apparatus, Micromeritics France S.A., Creil, France) in which the analysis gas flows into the sample tube and the balance tube at the same time. The rate of delivery of gas into the sample tube is controlled by the rate at which the sample can adsorb the gas onto the surface. The rate of flow into the balance tube is controlled to give the same pressure. A preliminary step consisted in removing water, carbon dioxide, and oxygen from the sample by heating and vacuum (40°C and 50 μm Hg). Each measurement was carried out on 8 g of grains that was ground as previously described, and two replicates were done.

In Situ Measurements. Ruminal starch degradation was determined by the in situ method as described previously (Philippeau and Michalet-Doreau, 1997). Three nonlactating Jersey cows fitted with a ruminal cannula received a diet of 70% hay and 30% concentrate. The concentrate was composed of 43% barley, 40% beet pulp, 10% soybean, 5% beet molasses, and 2% mineral-vitamin premix. Daily ration was 6 kg DM, given in two equal portions at 0800 and 1700. Cows were adapted to their diet for 4 wk before measurements. Each corn was ground through a 3-mm sieve in a hammer mill (Culatti, DFH48 type; Prolabo, Fontenay-sous-Bois, France). Approximately 3 g of ground grain was put into nylon bags (Ankom Co, Fairport, NY; pore size: 53 μm; internal dimensions: 5×10 cm) and introduced into the rumen at the same time just before the morning meal. They were removed after 3, 6, 9, 15, 24, and 48 h of incubation. Six measurements (two repetitions × three cows) were made for each incubation time. After removal, bags were rinsed in cold water, frozen, and washed in a washing machine with three successive 2-min washings. They were then dried at 80°C for 48 h and weighed. Starch content was determined on the incubation residues (Faisant et al., 1995). The proportions of particulate DM and starch that passed through the pores of the bag without being degraded were determined for each corn according to the method previously described (Philippeau and Michalet-Doreau, 1997). Three grams of DM was placed in nylon bags, immersed in 250 mL of a buffer solution at pH 6.9, and agitated for 2 h in a 39°C water bath. After removal, the bags were rapidly washed with distilled water. Lost particles were recovered from the solution by filtration (6 μm). The filters were dried at 80°C for 48 h and weighed, and the starch content was determined (Faisant et al., 1995). Duplicate samples underwent the complete procedure.

The degradation kinetics of DM and starch obtained for each corn cultivar and for each animal were fitted with an exponential model (1) without a lag time, except for two corns (numbers 9 and 12), and (2) with a lag time (θ):

$$(1)\ \text{Disappearance}\ (t) = a + b\ (1 - e^{-ct})$$
$$(2)\ \text{Disappearance}\ (t) = a + b\ (1 - e^{-c(t-\theta)})$$

The equations suppose three dietary fractions: one rapidly degradable in the rumen (a); another with a slower degradation (b) with speed reducing exponentially [exp(–ct)]; and the last one undegradable (Ind = 100 – a – b). The three parameters, a, b, and c and the lag time were estimated by an iterative least squares procedure of SAS (1988), and best fit values were chosen using the smallest sums of squares after convergence. Dry matter and starch degradabilities were calculated with the equations of Ørskov and McDonald (1979) and Dhanoa (1988), respectively, at a rumen outflow rate of .06 h^{-1} (Poncet et al., 1995).

Chemical Analyses. Dry matter (130°C, 24 h), starch with an enzymatic method (Faisant et al., 1995), and nitrogen with the Dumas method (AOAC, 1990) were determined. All chemical analyses were performed in duplicate.

Statistical Analysis. Dry matter and starch degradation data were evaluated with an analysis of variance using the GLM procedure of SAS (1988) in a factorial model with two main factors, animal and endosperm texture. Chemical and physical data were evaluated with an analysis of variance with one factor, the endosperm texture. Starch degradability was predicted with the multiple linear regression equations built using forward stepwise selection (SAS, 1988). The vitreousness, the 1,000-grain weight, and physical characteristics of grains were added one by one to the model until no remaining variable produced a significant (P = .10) F-statistic.

Results and Discussion

The characteristics of the 14 corns are described in Table 2. The vitreousness ranged from 38.5 to 79.1% and averaged 51.4 and 71.8% in dent and flint types, respectively. The 1,000-grain weight was higher for dent than for flint types, averaging 278 and 222 mg, respectively. There was also wide variation among corn types in chemical composition. Cornstarch content ranged from 60.1 to 72.0%, and CP content ranged from 8.7 to 13.5%. The average starch content was similar for dent and flint types (68.0 vs 67.1%), whereas the crude protein content was slightly lower for dent than for flint corns (10.7 vs 12.0%). The vitreousness was not related to the starch content but was linked by a linear relation (P < .05) to the CP content. The floury endosperm has a thinner protein matrix than the horny portion; this could result in a lower CP content in the floury than in the horny portion (Hamilton et al., 1951).

The dent and flint corns differed markedly in the proportion of coarse particles, apparent density, and specific surface area (Table 3). In agreement with Chandrashekar and Kirleis (1988) and Le Deschault de Monredon et al. (1996), the dent types were characterized by a smaller proportion of coarse particles (61.9 vs 69.6%) and inversely by a higher proportion of fine particles (15.6 vs 9.0%). These results confirm the empirical observation that a vitreous corn is hard (Shull et al., 1990; Li et al., 1996). In contrast to results reported by Szaniel et al. (1984), we did not find a close association (r^2 = .06) between the two hardness estimators (i.e., the proportion of coarse particles and the grinding energy). Our measurement of grinding energy was of little precision. The grinding energy ranged from 3.0 to 4.1 kWh/t, whereas the minimal variation in grinding energy that could be measured was .3 kWh/t. The apparent density was lower (P < .01) for dent than for flint

Table 2. Influence of corn grain texture on corn grain characteristics

	Dent			Flint				
Item	Mean	Minimum	Maximum	Mean	Minimum	Maximum	SEM	*P*-values[a]
Vitreousness, %	51.4	38.5	57.3	71.8	66.8	79.1	1.40	.0001
1,000-grain weight, g	278	223	336	222	210	270	10	.0208
Starch, % DM	68.0	62.6	71.8	67.1	60.1	72.0	.86	.6215
CP, % DM	10.7	8.7	12.6	12.0	10.8	13.5	.33	.0638

[a] Dent as flint.

Table 3. Influence of corn grain texture on physical characteristics of corn grain

	Dent			Flint				
Item	Mean	Minimum	Maximum	Mean	Minimum	Maximum	SEM	*P*-values
Grinding resistance, kWh/t	3.7	3.2	4.1	3.7	3.0	4.0	.03	.904
Particles, %								
Coarse	61.9	42.0	66.7	69.6	68.6	70.5	.46	.045
Intermediate	22.2	18.6	29.4	21.1	18.5	27.6	.24	.573
Fine	15.6	5.2	34.8	9.0	1.8	11.2	.49	.103
Mean particle size, μm	869.3	718.1	1,004.9	946.2	827.3	1,110.2	26.9	.183
Density, g/cm^3								
Apparent	1.29	1.17	1.32	1.36	1.32	1.37	.0	.007
True	1.40	1.38	1.42	1.40	1.38	1.41	.0	.302
Specific surface area, m^2/g	.13	.07	.22	.07	.05	.101	.0	.006

corns, 1.29 vs 1.36 g/cm^3, and was strongly correlated with the grain vitreousness ($r^2 = .71$). These results confirm the close correlation between grain density and vitreousness as shown by Chandrashekar and Kirleis (1988) and Mestres et al. (1991). The wide difference in density among dent and flint types could be explained not only by differences in biochemical composition, but also by differences in the amount of void spaces within the endosperm and consequently by the ratio of horny to floury endosperms. Horny endosperm is very dense, whereas floury endosperm is full of microfissures or void spaces (Watson, 1987). Accordingly, after grinding, the true density did not vary between dent and flint types because there were no air void spaces left. The specific surface area corresponded to the theoretical surface area available to microbial attack by enzymatic protein. The ground corn specific surface area was low, averaging .1 m^2/g. This result agreed with surface area determined for corn grain by Melcion and de Monredon (1987), and it was slightly lower than that of ground wheat grain (Chesson et al., 1997). The distribution of pores would be determined in wheat whole grain by the bran fraction instead of by the endosperm. But, for corn grain, the specific surface area of the whole grain depended greatly on the texture of the endosperm. It was higher for dent than for flint type, .13 and .07 m^2/g, respectively, and it was negatively correlated to vitreousness ($r^2 = .63$).

The effective DM degradability averaged 50% and ranged from 39.7 to 71.5%. An increase in DM degradability was linked to increases in the rapidly degradable fraction and the degradation constant rate (Table 4). Ruminal starch degradability averaged 55.1% (SE = 1.4) and was characterized by a wider range of variation (40.6 to 77.6%) than ruminal DM degradability (Table 5). The undegradable starch fraction was nil for each corn cultivar. A higher starch degradability was mainly due to a higher rapidly degradable fraction (5.3 to 36.3%) to the detriment of the slowly degradable fraction (94.7 to 63.7%), and to a higher degradation constant rate (.036 to .111 h^{-1}). Two corns (numbers 9 and 12) were exceptions; the ruminal starch degradation began with a lag time of 12.2 and 7.8 h, respectively. As shown by Denham et al. (1989), including a lag time in the model used to describe ruminal starch disappearance led to a larger rapidly degradable starch fraction and to a smaller slowly degradable starch fraction. In this study, the increase in the rapidly degradable starch fraction was emphasized for these two corns because the degradation constant rate was higher than that of the other flint varieties. The mean ruminal starch degradability was slightly lower than results obtained on 3-mm ground samples (Cerneau and Michalet-Doreau,1991), but the range of variation was wider than data reported on corn grain (Nocek and Tamminga, 1991). The extent of the variation between

Table 4. Influence of corn grain texture on ruminal DM degradation of corn grain

	Dent			Flint				
Item	Mean	Minimum	Maximum	Mean	Minimum	Maximum	SEM	*P*-values
Rapidly degradable fraction, %	23.8	17.8	38.1	13.6	11.3	16.7	1.4	.0001
Slowly degradable fraction, %	76.2	61.9	82.2	86.4	83.3	88.7	1.4	.0001
Degradation constant rate, h^{-1}	.045	.038	.071	.030	.028	.032	.002	.0001
Effective degradability, %	55.8	51.9	71.5	42.3	39.7	45.3	1.3	.0001
Particle losses, %[a]	14.2	8.1	29.6	7.7	5.4	18.3	1.5	.0583

[a] Percentage of DM initially incubated.

Table 5. Influence of corn grain texture on ruminal starch degradation of corn grain

	Dent			Flint				
Item	Mean	Minimum	Maximum	Mean	Minimum	Maximum	SEM	*P*-values
Rapidly degradable fraction, %	26.6	19.4	36.3	19.4	5.3	32.3	2.2	.0072
Slowly degradable fraction, %	73.4	63.7	80.7	80.6	67.7	94.7	2.2	.0072
Degradation constant rate, h^{-1}	.059	.048	.111	.039	.034	.050	.004	.0003
Effective degradability, %	61.9	55.1	77.6	46.2	40.6	50.5	1.5	.0001
Particle loss, %[a]	12.9	7.7	26.6	5.7	4.1	17.9	1.5	.022

[a] Percentage of starch initially incubated.

corn cultivars was also much greater than that reported on other cereals: 15 points for barley (G. R. Khorasani, personal communication) and less than 10 points for sorghum (Streeter et al., 1990a,b). In this study, the extent of the variation was slightly higher than that reported between corn and barley (Cerneau and Michalet-Doreau, 1991). The DM degradation traits and effective degradability were strongly linked to the starch degradation traits and effective degradability (Table 6). Ninety-eight percent of the variation in the extent of starch degradability accounted for that of DM degradability.

The effective starch degradability was higher for dent than for flint corns, averaging 61.9 and 46.2%, respectively. These two corn types differed in vitreousness, averaging 51.4 and 71.8%, respectively (Table 2). The increase in the effective starch degradability was due to an increase in the rapidly degradable starch fraction and to an increase in the degradation constant rate. The variation in the rapidly degradable fraction might be related to the difference in the proportion of particulate starch losses. The particulate starch losses were higher ($P < .02$) for dent than for flint corns (Table 2), but they remained low for the two types of corn: less than 17.8% starch initially incubated except for one corn (26.6% starch initially incubated) (Table 5). Within each type, the extent of the variation in ruminal starch degradation was wider for dent than for flint corns (22.5 and 9.9 points), and it was linked to the wider range of variation of the vitreousness for dent than flint corns (18.8 and 12.3 points, respectively). These results agreed with previous results that reported large variations in ruminal starch degradation between dent and flint corns in in vitro (Ladely et al., 1995; Opatpatanakit et al., 1994) and in situ (Michalet-Doreau and Champion, 1995) starch digestion studies.

Ruminal starch degradability and physical characteristics varied widely among corn cultivars. An examination of the relationships between these grain characteristics and ruminal starch degradability revealed potential for predicting ruminal starch digestion of cornstarch for ruminants. As we have shown for immature corn grain (Philippeau and Michalet-Doreau, 1997), 88.5% of the variation in ruminal starch degradability was associated with vitreousness, and this endosperm characteristic was a good predictor of ruminal starch degradability (root mean square error [**RMSE**] = .9). The most accurate prediction for ruminal starch degradability was provided by taking into account two additional traits: apparent grain density and 1,000-grain weight ($R^2 = .97$; RMSE = .4). In practice, vitreousness was very time-consuming to measure. Thus, we sought predictive models in which the vitreousness need not be considered. Apparent density was strongly related to grain vitreousness. This predictor of ruminal starch degradability led to a high coefficient of determination, but the RMSE was slightly lower ($R^2 = .81$; RMSE = 1.1). A better prediction was obtained by combining the apparent density with the 1,000-grain weight ($R^2 = .91$; RMSE = .8). These two measurements were not time-consuming.

Table 6. Coefficient of correlation between ruminal DM and starch degradation

	Starch			
Variable	Rapidly degradable fraction, %	Slowly degradable fraction, %	Degradation constant rate, /h	Effective degradability, %
Rapidly degradable DM fraction, %	.74**	.33	.83**	.88**
Slowly degradable DM fraction, %	.39	.95**	.33	.63*
DM degradation constant rate, h^{-1}	.61*	.46	.95**	.90**
Effective DM degradability, %	.72**	.66**	.87**	.99**

*$P < .05$.
**$P < .01$.

Implications

The variability in ruminal starch digestion of corns differing in endosperm texture indicates that genetic selection holds promise for manipulating the site and extent of starch digestion in ruminants. The site of starch digestion alters the nature of the end products of digestion (i.e. volatile fatty acids in the rumen and hindgut and glucose in the small intestine) and, in this respect, the efficiency of their metabolic utilization by the ruminant. In this trial, in situ ruminal starch degradability was closely linked to ruminal dry matter degradability. But, this relationship should be confirmed with a great number of corns. Moreover, ruminal starch degradability of corn grain can be accurately predicted by the grain vitreousness and rapidly measurable physical traits.

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