

ANALYSIS AND PREDICTION OF UNREACTED STARCH CONTENT IN CORN USING FT-NIR SPECTROSCOPY

B. M. Plumier, M. C. Danao, V. Singh, K. D. Rausch

ABSTRACT. *When corn is processed in a conventional dry-grind ethanol process, a portion of the corn starch is not readily converted into ethanol. The amount of unconverted, or unreacted, starch varies according to several factors, including storage time and processing conditions. The current method for determining the amount of unreacted starch is based on an enzyme assay, which is time-consuming and does not lend itself to on-line measurements of corn in processing plants. A rapid method for determining the unreacted starch in corn would be advantageous so that the mix of enzymes and processing conditions could be adjusted to ensure maximum ethanol yield. In this study, we demonstrated the feasibility of using Fourier transform near-infrared (FT-NIR) spectroscopy in developing predictive models of unreacted starch in corn. FT-NIR spectra of corn starch blends and ground corn from 4000 to 10000 cm^{-1} were calibrated against unreacted starch content, determined enzymatically, using various spectral preprocessing techniques such as multiplicative scatter correction (MSC), Savitzky-Golay (SG) derivative algorithms, and partial least squares regression (PLSR). Results showed that the unreacted starch content in blends can be predicted with a low root mean square error of prediction (RMSEP) ranging from 1.29% to 1.95%, a coefficient of regression (R^2) of 0.97 to 0.98, and a ratio of performance to deviation (RPD) of 4.82 to 7.28. PLS regression models for unreacted starch content in dry and wet ground corn were equally promising, with low RMSEP values of 1.13% to 2.23%, R^2 values of 0.83 and 0.94, and RPD values of 1.55 to 2.16. These models are a valuable tool for high-throughput monitoring of unreacted starch during corn storage, handling, and processing.*

Keywords. *Bioprocessing, Dry-grind ethanol, Near-infrared spectroscopy, Partial least squares.*

The most common method used for converting corn into ethanol is the dry-grind process, which is responsible for 86% of domestic ethanol production (Mueller, 2010). Because yeast cannot utilize starch directly, the production of ethanol from corn requires hydrolysis of starch to glucose, which is then fermented by yeast to ethanol. Unconverted starch (sometimes called unreacted, residual, or resistant starch) is eventually recovered in distillers dried grains with solubles (DDGS). DDGS, a coproduct of the process, can contain up to of 5% starch (Belyea et al., 2004; Kim et al., 2008). Since 3.8 L (1 gal) of ethanol produces 2.7 kg of DDGS (Szulczyk et al., 2010), this means that for the 43.2 billion L (11.4 billion gal) of ethanol produced in the U.S. in 2011, 1.5 billion kg of starch was unconverted, or unreacted. This represents inefficiencies in the dry-grind ethanol process, and reducing the amount of unreacted starch is particularly important to the economics of the ethanol industry. In order for ethanol plants to maximum their yields and adjust their choices of enzyme mixtures,

yeast strains, and processing conditions efficiently, it would be beneficial to develop a high-throughput procedure for monitoring unreacted starch in corn during storage and handling, and prior to conversion.

Starch makes up 70% to 73% of the corn kernel dry weight (Srichuwong and Jane, 2011). It is known that enzymatic digestibility of raw starch granules varies among corn varieties (Perera et al., 2001) due to the differences in amylose content and branch-chain length of the amylopectin (Ji et al., 2003; Jane, 2009). Resistant, or unreacted, starch content in corn is highly correlated with amylose level (Berry, 1986). Yangcheng et al. (2013) reported an average starch-to-ethanol conversion efficiency of 93.0% for waxy corn, which has a low amylose:amylopectin ratio, and this value was greater than that of normal corn (88.2%). The physical damage in starch granules that results from dry-grinding corn can also lead to greater digestibility of the starch (Karkalas et al., 1992; Tester et al., 1994). Since yeast cannot utilize starch directly, starch is first hydrolyzed to glucose; often, however, not all of the starch is converted to sugars. The amount of unconverted starch recovered depends on the type of raw starch in the corn and the process parameters, such as temperature, pH, enzyme activity, and duration of hydrolysis (Berry, 1986; Sharma et al., 2010).

Currently, unreacted starch content in corn is determined using enzymatic colorimetric assays (AOAC, 2003; Hollmann et al., 2013). Vidal et al. (2009) developed a procedure that involved fermenting the corn sample and using a D-glucose assay kit to estimate the glucose content in the ground corn and back-calculating the unreacted starch.

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The authors are **Benjamin M. Plumier, ASABE Member**, Graduate Student, **Mary-Grace C. Danao, ASABE Member**, Assistant Professor, **Vijay Singh, ASABE Member**, Professor, and **Kent D. Rausch, ASABE Member**, Associate Professor, Department of Agricultural and Biological Engineering, University of Illinois, Urbana, Illinois. **Corresponding author:** Mary-Grace C. Danao, 376B AESB, MC-644, 1304 West Pennsylvania Ave., Urbana, IL 61801; phone: 217-244-3925; e-mail: gdanao@illinois.edu.

Dry-grind ethanol plants can also determine the unreacted starch content after fermentation by analyzing the DGGS or comparing predicted ethanol yields to actual ethanol yields. All of these methods can take several hours or days to complete and usually require access to a wet chemistry lab and expensive analytical equipment.

As with most grain, forage, and feed analyses, wet chemistry assays may be substituted with near-infrared (NIR) spectroscopy analyses, provided that calibration models are developed and validated. NIR analyses have long been used in the food and pharmaceutical industries because they offer high accuracy, precision, and timely analyses of raw and processed materials. In most NIR applications, little sample preparation is required, and the sampling procedure is noninvasive and nondestructive, allowing for on-line processing implementation (Blanco and González Bañó, 2003; Tikuisis et al., 1993).

NIR spectra of ground corn typically show strong absorption bands of sugars, proteins, oils, and starches, which usually overlap with water absorption bands if the corn is not sufficiently dried prior to scanning (table 1). Glucose, maltose, and dextrans overlap between 4255 and 4762 cm^{-1} and are associated with C-O and O-H deformation and O-H and C-C stretching vibrations (Noah et al., 1997; Blanco and González Bañó, 2003). Starch has absorption bands at 4394 to 4762 cm^{-1} and 6250 to 7143 cm^{-1} , with the latter corresponding to the first overtone of hydroxyl groups (Noah et al., 1997).

Some studies have reported the use of infrared spectroscopy in starch and sugar analyses of corn and corn products. Ruminant digestive resistant starches in variety corn, wheat, and potato samples were modeled using mid-infrared spectroscopy with a mean square prediction error of 0.0076% and $R^2 = 0.995$ (Udén, 2009). Hydrolysates

from the dry-grind ethanol process were measured for sugar concentrations using Fourier transform near-infrared (FT-NIR) spectroscopy with less than 5% error (Blanco and González Bañó, 2003). Paulsen et al. (2010) demonstrated that residual starch content in DDGS can be predicted with $R^2 = 0.81$ to 0.87. Garcia-Rosas et al. (2009) found that infrared spectra demonstrated the retrogradation of stored starch in corn tortillas by changes in the shapes and intensities of the characteristic peaks at 1022 and 1047 cm^{-1} .

In this study, partial least squares regression (PLSR) models of unreacted starch content in starch blends and ground corn were developed based on their FT-NIR spectra. Dry blends of digestible and digestively resistant starch were first scanned and modeled to determine which wavebands of the FT-NIR spectra were most responsive to changes in unreacted starch content. The FT-NIR spectra of dry ground corn were compared to the FT-NIR spectra of the dry starch blends to ensure that the previously identified wavebands were not masked by other constituents in the ground corn and would remain prominent in the regression models. However, for on-line monitoring of ground corn in a dry-grind ethanol plant, it will be most advantageous to develop predictive models that can be used with wet ground corn. Therefore, the FT-NIR spectra of wet ground corn were also collected, compared to the spectra of the dry starch blends and dry ground corn samples, and modeled using PLSR.

MATERIALS AND METHODS

SAMPLE PREPARATION AND UNREACTED STARCH CONTENT DETERMINATION

Starch blends (0% to 33% unreacted starch content) were prepared by mixing weight proportions of Hi-maize resistant starch (Honeyville, Brigham City, Utah) with soluble and hydrolyzed starch (Product No. S516, Fisher Scientific, Fair Lawn, N.J.). Honeyville's product contains Hi-maize 260 (Ingredion, Inc., formerly Corn Products/National Starch, Bridgewater, N.J.) resistant starch that has been isolated from high-amylose corn hybrids produced through traditional plant breeding and contains 33% digestible, or glycemic, starch. Based on weight ratios, a total of 56 starch blend samples were prepared with unreacted starch contents ranging from 0% to 33%; three samples with three replications were also used to verify the unreacted starch content using the same procedure used for corn harvested in 2011. The starch blends were subjected to a liquefaction and saccharification procedure, washing out the glucose by centrifugation, and measuring the remaining starch in the hydrolysate. Results from the verification test are shown in the following section.

Pioneer P1395R yellow dent corn was harvested from the Agricultural and Biological Engineering Farm in Urbana, Illinois, in October 2011, and the same hybrid was provided by Dupont Pioneer Hi-Bred International (Johnston, Iowa) directly in October 2012. Wet corn flour samples were prepared by periodically taking 200 to 300 g samples from storage and grinding the samples in a hammer mill using a 0.5 mm sieve (fig. 1). A 10 g ground corn subsample was used to determine the ground corn moisture content by gravimetric method and drying at 135°C for 2 h (AACC, 2000).

Table 1. NIR wavebands of different constituents in ground corn.

Wavenumbers (cm^{-1})	Assignment	References
4301-4415	Starch yields for wet milling	Wehling et al. (1993)
4283-4329	C-H bending and C-H stretching, carbohydrate IV	Hódsági et al. (2012), Hao et al. (2012)
4367-4396	O-H stretching and C-C stretching, carbohydrate III	Hódsági et al. (2012), Wehling et al. (1993)
4587-4673	NH band indicating protein	Paulsen et al. (2003)
4864-4873	Protein	Hao et al. (2012), Paulsen et al. (2003)
5250	Amylose and amylopectin helix structures	Lindberg and Kaila (1980)
5225-5319	Starch	Hao et al. (2012)
4274, 4333, 5814, 5682, and 8652	CH_2 bands indicating fat or oil	Paulsen et al. (2003)
5800	C-H stretching, first overtone	Hao et al. (2012)
6289-6349	O-H stretching, first overtone, carbohydrate I	Hódsági et al. (2012)
6684	NH band indicating protein	Paulsen et al. (2003)
8889-9524	Starch yields for wet milling; C-H stretching, C-C stretching associated with lipids and other hydrocarbons	Wehling et al. (1993)
5155, 6897, 8403, and 10309	Water absorption bands	Ben-Gera and Norris (1968), Paulsen and Singh (2004)
10823	Extractable starch	Paulsen and Singh (2004)

For corn harvested in 2011, ground corn was first liquefied for 1 h with Liquozyme SC alpha amylase (Novozymes, Franklinton, N.C.) for 72 h at pH 5.7 and 85°C, followed by saccharification with Spirizyme Ultraglucoamylase (Novozymes) for 72 h at pH 5.0 and 65°C. The samples were then washed with deionized water three times in a centrifuge set at 500 rpm for 20 min to remove glucose from the samples. The samples were dried at 49°C overnight and treated with either deionized water (blank samples) or 0.4 M hydrochloric acid (test samples) to break apart the remaining starch molecules. All blank and test samples were diluted 10× using deionized water, and the unreacted starch content was determined using a glucose-oxidase-peroxidase (GOPOD) colorimetric assay (Megazyme International, Wicklow, Ireland). Estimates of the glucose content obtained were used to calculate the original unreacted starch content remaining after saccharification based on a procedure developed by Vidal et al. (2009). This procedure relied on dilute hydrochloric acid

to measure the enzymatic unreacted starch; acid-resistant starch was not measured in this study.

For corn harvested in 2012, the unreacted starch assay was modified by conducting a simultaneous saccharification and fermentation with Spirizyme Ultraglucoamylase at pH 4.5 and 32°C for 72 h, which is more representative of the procedures used in modern dry-grind ethanol facilities and offered complete removal of fermentable sugars in corn samples. Ethanol was removed by distillation at 90°C for 3 h and dried at 49°C for 72 h. The GOPOD colorimetric assay was used to determine the amount of glucose remaining in the sample and converted to unreacted starch content.

Note that there were three measures of unreacted starch content for each of the three replications of dry ground corn. These values were averaged and assigned to the FT-NIR spectra collected for the wet ground corn prior to liquefaction.

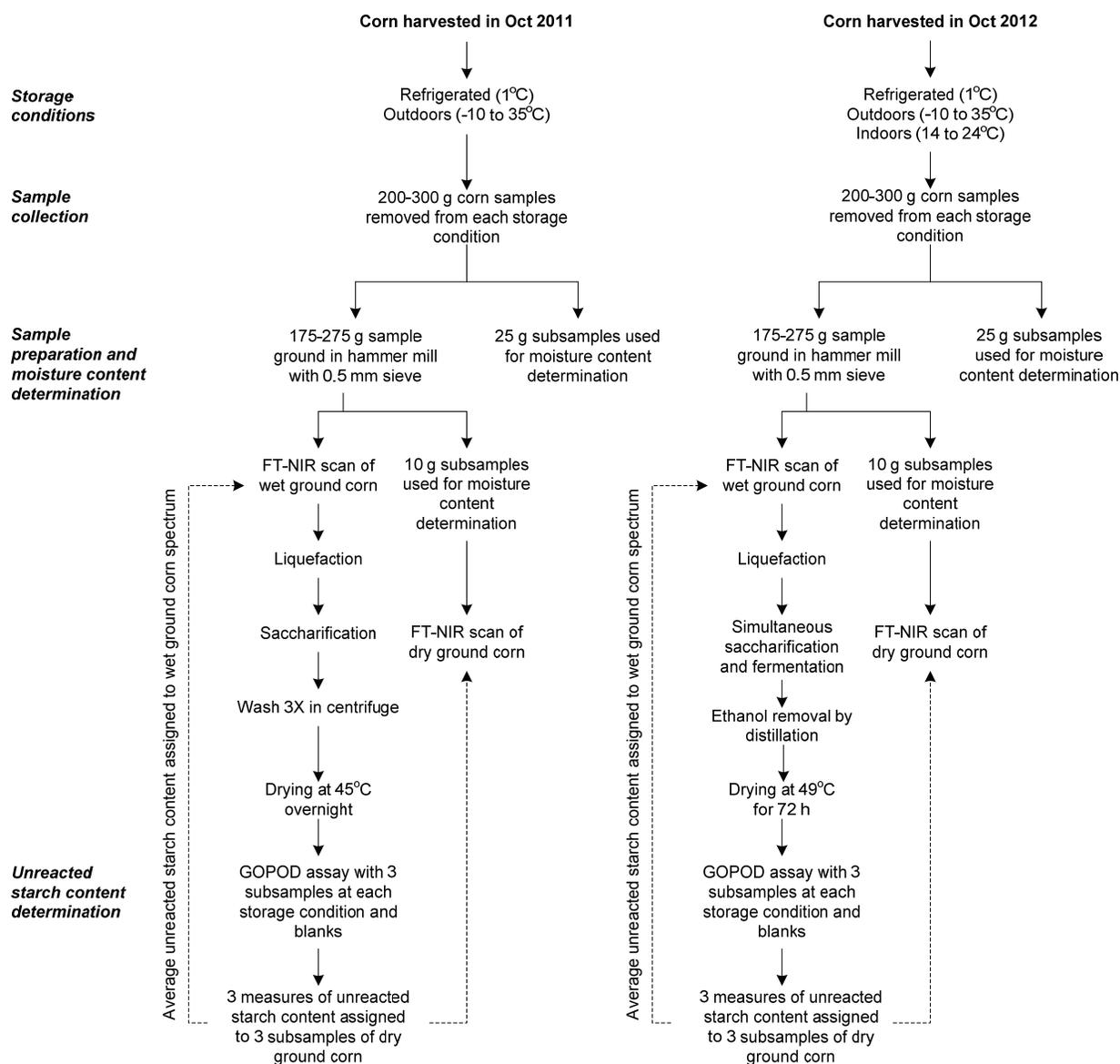


Figure 1. Preparation of dry and wet ground corn used in developing PLSR models based on FT-NIR spectroscopy. The same liquefaction, saccharification, centrifugation, and GOPOD assay used for corn harvested in 2011 was used to verify the unreacted starch content in the dry starch blends.

VERIFICATION OF UNREACTED STARCH CONTENT

Three samples with three replications each of starch blends with unreacted starch content ranging from 16% to 34% were prepared and tested using the same liquefaction, saccharification, centrifugation, and colorimetric GOPOD assay used with the 2011 harvested corn. Results of the colorimetric assay were misleading, as nearly all of the digestible starch was converted to glucose and removed, leaving nothing but the unreacted starch. The calculations led to unreacted starch contents of 80% and greater. A small amount of glucose, ranging from 12% to 17%, was found to remain after centrifuge washing. The percentage of non-starch in the hydrolysate ranged from 4% to 9%. However, the amount of solid hydrolysate apparent in the samples varied greatly after centrifuge washing, and a strong correlation was found between the theoretical resistant starch content and the weight of the hydrolysate, with $R^2 = 0.9982$ (fig. 2). This test demonstrated that the commercially available digestive resistant starch was an adequate analog for enzymatically resistant starch, and the starch blends produced could be used in developing FT-NIR techniques for screening unreacted starch in corn.

SCANNING, PREPROCESSING, AND ANALYSES OF FT-NIR SPECTRA

An FT-NIR spectrophotometer (Spectrum One NTS,

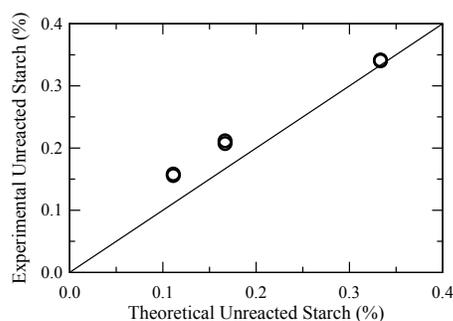


Figure 2. Comparison between experimental to theoretical unreacted starch content in starch blends. The unreacted starch content was measured by weight after centrifuge washing.

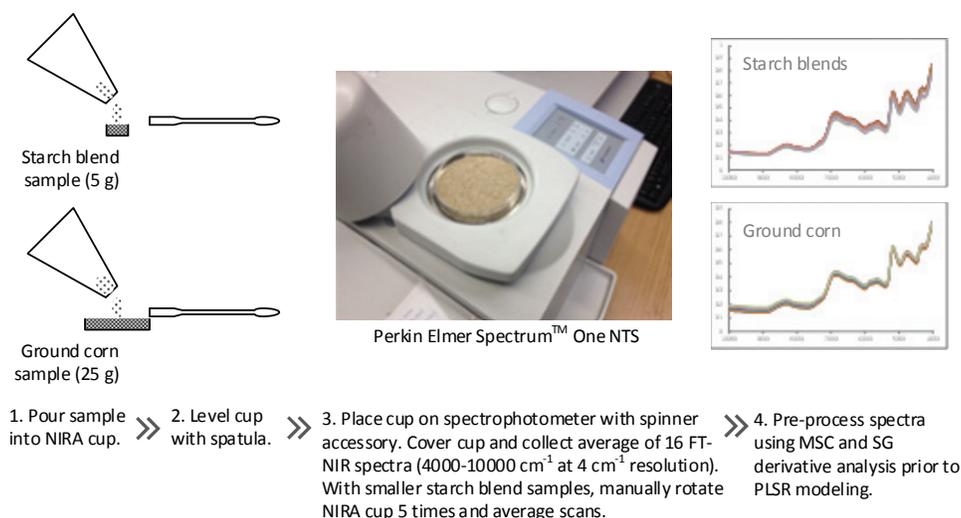


Figure 3. Collection of FT-NIR spectra of starch blends and ground corn samples.

Perkin Elmer, Waltham, Mass.) was used to scan dry starch blends and dry ground corn samples from 4000 to 10000 cm^{-1} with a spectral resolution of 4 cm^{-1} . Approximately a 5 g subsample was poured into a near-infrared reflectance accessory (NIRA) cup, leveled with a spatula, and scanned 16 times, manually rotating the cup 30° to 45° clockwise between each scan. This procedure was repeated five times with separate subsamples, and the resulting spectral scans were averaged. For the wet ground corn samples, approximately 25 g was poured into a NIRA cup and leveled with a spatula. The cup was placed on a spinner attachment, and the spectrophotometer was set to take an average spectral scan from 16 readings (fig. 3). Unscrambler (version 10.2, Camo Software, Inc., Woodbridge, N.J.) was used to preprocess and analyze the spectral data with multiplicative scatter correction (MSC) or Savitzky-Golay (SG) derivative filtering and build, validate, and evaluate PLSR models based on number of factors, coefficient of determination (R^2) between the predicted and reference unreacted starch values, root mean square error of prediction (RMSEP), and ratio of standard error of performance to standard deviation (RPD). An RPD value greater than 8 is generally considered good for process control, development, and applied research, 5 to 8 indicates that the model is acceptable for quality control, and models with RPD values between 2.5 to 5.0 are acceptable for screening samples (AACC, 1999).

RESULTS AND DISCUSSION

UNREACTED STARCH CONTENT IN STARCH BLENDS AND GROUND CORN

The starch blends had unreacted starch contents ranging from 0% to 33.3% resistant starch, with a mean of 16.69%. There were 36 samples in the calibration set, 14 samples were used in the validation set, and six samples were used in the test set. The dry ground corn regression models were made using 100 samples of corn: 72, 20, and 8 samples were used for the calibration, validation, and test sets, respectively. Unreacted starch contents in the dry ground corn ranged from 5.1% to 16.6%, with a mean of 11.9%.

Similarly, 49 samples of corn were used to develop the PLS models for the wet ground corn: 36 samples for calibration, 10 samples for validation, and 3 samples for the test set. Unreacted starch contents in the wet ground corn ranged from 1.6% to 15.1%, with a mean of 10.1%.

PREPROCESSING OF FT-NIR SPECTRA

The FT-NIR spectra of the starch blends and ground corn samples showed prominent peaks, both crests and troughs, at around 5250 cm^{-1} and 7000 cm^{-1} (fig. 4). The region around 5250 cm^{-1} has been noted for O-H stretching and O-H deformation of starch components, specifically amylose and amylopectin helix structures. This region was used as the basis for measuring the stability of the helical structures in the gel state in the presence of water and dimethylsulfoxide (Lindberg and Kiala, 1980). While all the samples showed the same FT-NIR spectral trends at 5250 cm^{-1} , the heights and slopes were more prominent in the dry ground corn samples. The peak at 7000 cm^{-1} has been reported as an important region for starch and protein (Noah et al., 1997), but it is also near an overtone of water absorption (Paulsen and Singh, 2004). This peak tended to be higher in the wet ground corn samples, so care should be taken that resulting PLSR not have high regression coefficients in this region.

FT-NIR scans of ground waxy corn and high-amylose corn were also conducted for reference, as the difference in amylose and amylopectin ratios would mean that those samples represent the opposite ends of theoretical unreacted starch content in corn (fig. 5). From 4000 to 6000 cm^{-1} , the peaks and troughs for waxy corn were steeper than for high-amylose corn, and the absorbance values were higher for waxy corn in both the peaks and troughs. Since higher

amylose contents are known to correlate with an increase in unreacted starch content, one would expect higher unreacted starch contents to correlate with lower absorption values in this region. This trend held true for most samples; in the 4000 to 4500 cm^{-1} region, absorbance values increased with decreasing unreacted starch content.

The baseline shifts in the raw spectral data (figs. 4 and 5) often do not represent compositional information but likely represent common sources of error in sample preparation or scattering due to particle size distribution during scanning. To remove the baseline shifts and other errors in the spectra, MSC and SG first and second derivatives were used. MSC corrected simple baseline shifts without over-processing, or changing, the appearance or location of the peaks and troughs as much as the derivative-based SG technique did. During SG derivative preprocessing, the number of smoothing points used changed how severely the spectra were processed. Too few points resulted in noisy, undersmoothed curves, whereas too many points resulted in oversmoothed curves. Oversmoothed curves eliminated spectral noise effectively but also tended to eliminate small absorption bands, or features, that were important indicators of unreacted starch.

PLSR MODELING

PLSR models using MSC, SG first and second derivatives, and a combination of MSC with SG second derivative preprocessed spectral data were developed for starch blends and ground corn (table 2). Models developed for the starch blends outperformed models developed ground corn, with R^2 values greater than 0.97, RPD values up to 7.28, and 95% explained variance. The best model involved a combined MSC and SG second derivative preprocessing,

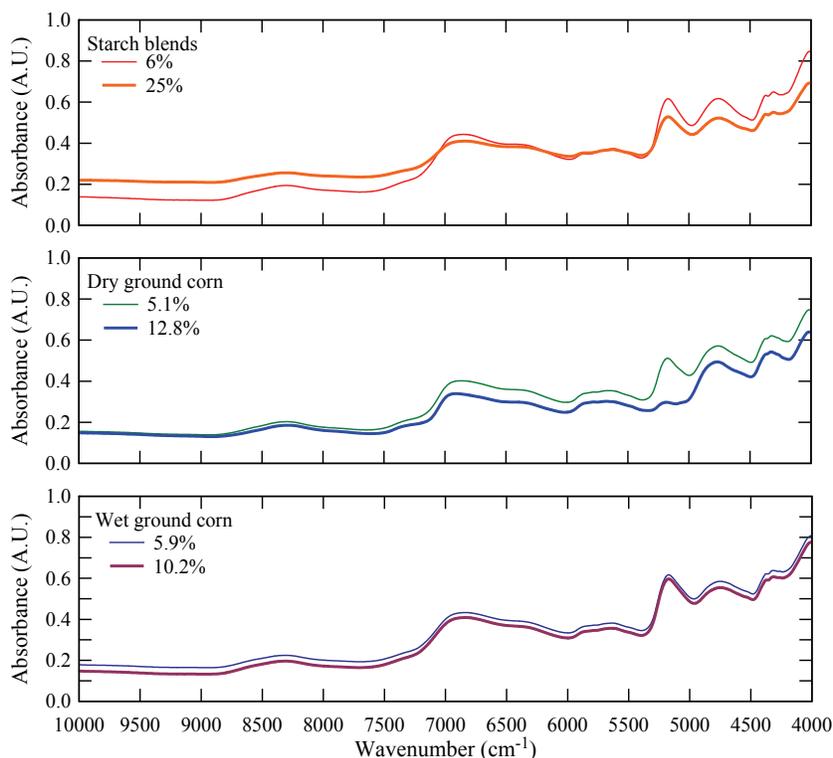


Figure 4. FT-NIR spectra of select starch blends and ground corn samples.

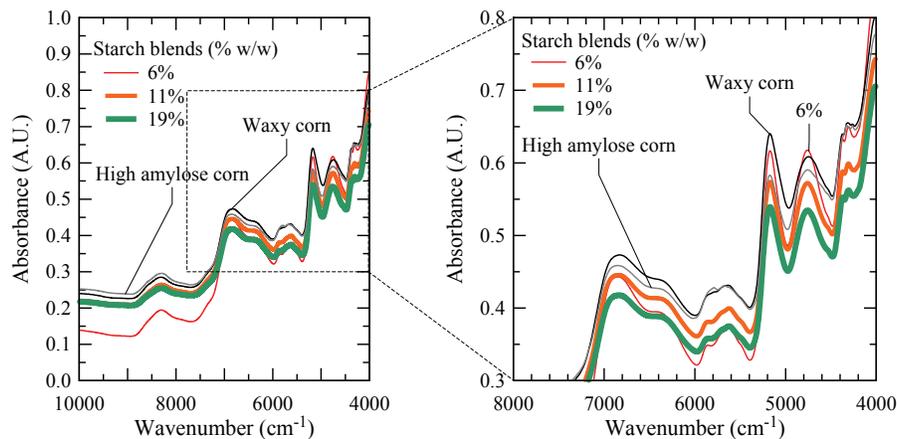


Figure 5. FT-NIR spectra of select starch blends with different unreacted starch contents (% w/w) and ground samples of waxy and high-amylose corn.

although results of the model using MSC preprocessed spectra were similar. The PLSR models for starch blends were expected to yield better results than the models for ground corn, as these samples did not have large amounts of the other constituents in corn, such as proteins, lipids, and moisture, which could mask the absorption bands indicating unreacted starch. The calibration and validation sets of the starch blends also had a wider range compared to the ground corn samples.

The best PLSR model for dry ground corn was also based on the combined MSC and SG second derivative preprocessed spectra. It had 85% explained variance, $R^2 = 0.86$, and $RPD = 2.07$. This model was based on the highest number of calibration and validation data sets, although the range of

unreacted starch contents was narrow, from 5.1% to 16.6% (fig. 6). This model could be improved with the addition of more ground corn samples, either of different varieties or stored for different time periods and conditions, and with a more accurate procedure for measuring leftover glucose in the DDGS other than the colorimetric GOPOD assay, such as high-performance liquid chromatography (HPLC).

The best PLSR model for wet ground corn was based on SG second derivative preprocessed spectra. It had 85% explained variance, $R^2 = 0.83$, and $RPD = 2.16$. This was better than the PLSR model developed for dry ground corn despite the fact that water would have interfered and masked important absorption bands in the spectra. One possible explanation for this was that the unreacted starch

Table 2. PLSR models of unreacted starch content in starch blends and ground corn samples.

Samples	Starch Blends (<i>N</i> , range, mean \pm SD) ^[a]				Dry Ground Corn (<i>N</i> , range, mean \pm SD) ^[a]				Wet Ground Corn (<i>N</i> , range, mean \pm SD) ^[a]			
	MSC	SG-1	SG-2	MSC+	MSC	SG-1	SG-2	MSC+	MSC	SG-1	SG-2	MSC+
Calibration	36, 0.0% to 33.3%, 16.6% \pm 10.1%				72, 5.1% to 16.6%, 12.0% \pm 2.2%				36, 1.6% to 15.1%, 9.8% \pm 3.9%			
Validation	14, 4.0% to 30.0%, 16.2% \pm 7.6%				20, 5.2% to 15.3%, 11.9% \pm 2.6%				10, 6.0% to 14.0%, 10.7% \pm 2.5%			
Test	6, 6.0% to 33.3%, 18.1% \pm 9.8%				8, 5.3% to 14.1%, 10.8% \pm 3.0%				3, 7.1% to 12.9%, 10.4% \pm 3.0%			
Data Preprocessing ^[b]	MSC	SG-1	SG-2	MSC+	MSC	SG-1	SG-2	MSC+	MSC	SG-1	SG-2	MSC+
No. of factors	3	2	2	2	4	4	2	2	3	4	3	3
No. of points in SG calculation	-	13	31	31	-	13	21	21	-	11	51	51
R^2 Values												
Calibration	0.97	0.97	0.98	0.98	0.41	0.83	0.83	0.86	0.70	0.93	0.83	0.94
Validation	0.96	0.93	0.93	0.97	0.49	0.64	0.75	0.80	0.19	0.38	0.54	0.47
Test	0.97	0.98	0.98	0.97	0.30	0.33	0.42	0.38	0.99	0.97	0.99	0.75
RMSE ^[c] (%)												
Calibration	1.81	1.63	1.44	1.23	1.77	0.89	0.89	0.82	2.14	1.01	1.59	0.96
Validation	1.51	1.94	1.95	1.29	1.29	1.51	1.26	1.13	2.23	1.95	1.67	1.81
Test	4.02	4.50	4.38	2.67	2.67	2.82	2.83	2.49	1.81	1.28	0.80	1.43
RPD ^[d]	6.22	4.84	4.82	7.28	1.81	1.55	1.86	2.07	1.61	1.85	2.16	1.99

^[a] Number of samples (*N*), range of minimum to maximum unreacted starch contents (%), and mean unreacted starch content \pm standard deviation (%) of the data set.

^[b] MSC = multiplicative scatter correction, SG-1 = Savitzky-Golay first derivative calculation using a second-order polynomial, SG-2 = Savitzky-Golay second derivative calculation using a second-order polynomial, and MSC + SG-2 = combined multiplicative scatter correction with a Savitzky-Golay second derivative calculation using a second-order polynomial.

^[c] RMSE = root mean square error: $RMSE = \sqrt{\sum_{i=1}^N (y_i - \hat{y})^2 / N}$, where y_i is the individual reference value (from wet chemistry assay), \hat{y} is the NIR predicted value from the PLS regression model, and *N* is the total number of samples.

^[d] RPD = ratio of standard error of performance to standard deviation ($RPD = \sigma_y / SEP$), where the standard deviation of the reference data is

$$\sigma_y = \sqrt{\sum_{i=1}^N (y_i - \bar{y}) / (N - 1)}, \bar{y} \text{ is the average reference value, and SEP is the standard error of prediction (using the validation set).}$$

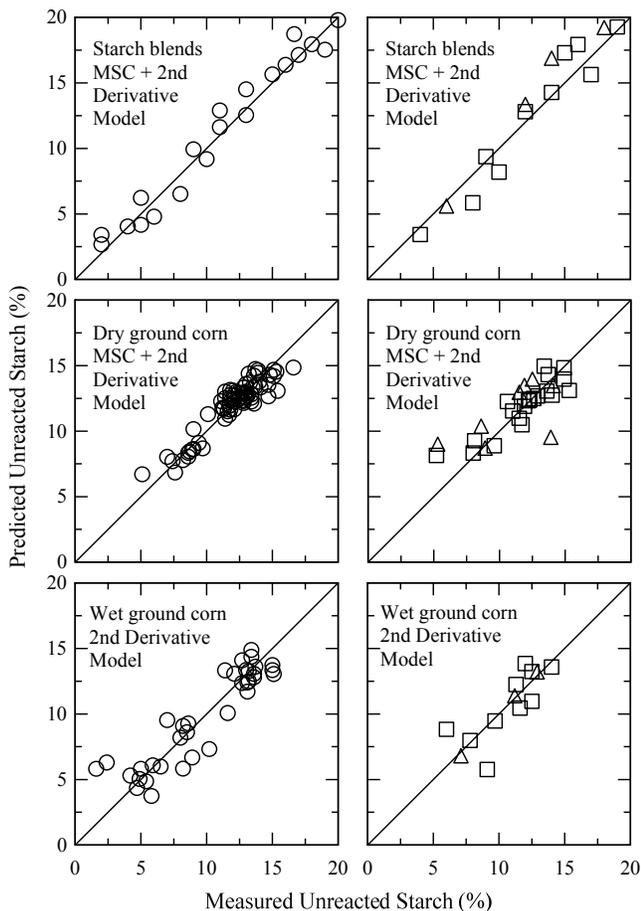


Figure 6. Comparison of predicted to measured unreacted starch content using the calibration (O), validation (□), and test (△) sets of starch blends and ground corn samples.

values for wet ground corn were based on the average of three replicates; for dry ground corn, measures were based on only one replicate. The calibration set for wet ground corn also had a wider range than for dry ground corn, from 1.6% to 15.1% with a standard deviation of 3.6%.

All of the models had high regression coefficients in the important wavebands of 4000 to 5000 cm^{-1} , 5250 cm^{-1} , and 7000 cm^{-1} for starch (fig. 7). However, the 7000 cm^{-1} region was likely attributable to protein and water content in the ground corn more than to unreacted starch content. The PLSR model for wet ground corn also relied on a peak around 5100 cm^{-1} which is a known water absorption band (Ben-Gera and Norris, 1968; Paulsen and Singh, 2004). Overall, since the crests and troughs of the regression coefficients coincided with the important wavebands for starch reported in the literature and found in the starch blends, with high absorption values at 5250 cm^{-1} , the resulting regression models were accurately predicting unreacted starch content in corn.

CONCLUSION

PLSR models using multiplicative scatter correction (MSC), Savitzky-Golay (SG) first or second derivative calculations, and combinations thereof to preprocess the FT-NIR spectra of starch blends and ground corn samples were developed. Models developed for the starch blends outperformed models developed for ground corn with R^2 values greater than 0.97, RPD values up to 7.28, and 95% explained variance. The best PLSR model achieved for dry ground corn was based on the combined MSC and SG second derivative preprocessed spectra, explaining 85% of the variance using two factors, with $R^2 = 0.86$ and $\text{RPD} = 2.07$. Similarly, the best PLSR model achieved for wet ground corn was based on SG second derivative preprocessed spectra, explaining 85% of the variance with three factors, with $R^2 = 0.83$ and $\text{RPD} = 2.16$. The RPD values suggest that FT-NIR spectroscopy is a promising technique for screening corn in dry-grind ethanol facilities, but it may be improved with the addition of waxy and high-amylose varieties of corn to widen the range of reference values in the calibration.

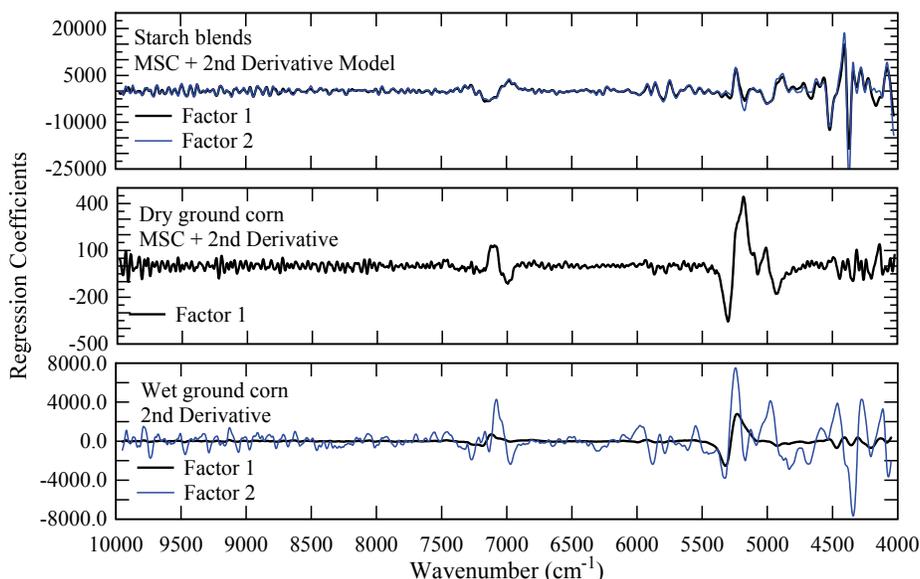


Figure 7. Regression coefficients of the best PLSR models developed for predicting unreacted starch content in starch blends and ground corn.

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