Compositional Variability Among Corn Stover Samples

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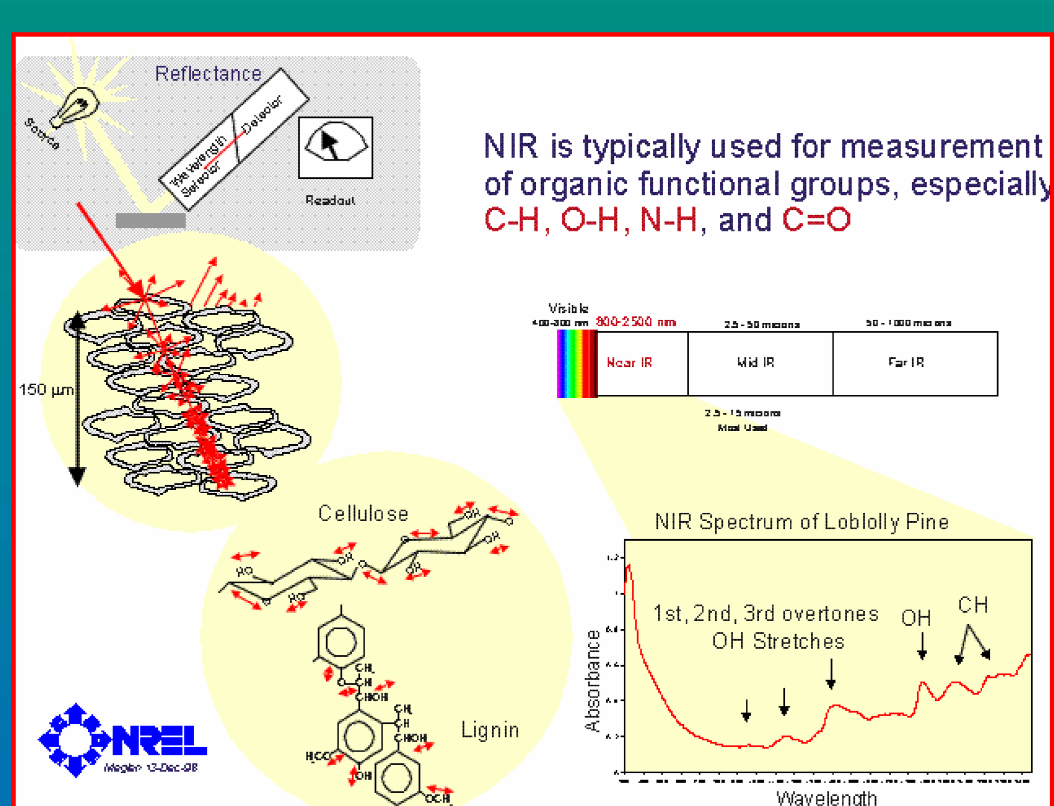
1. Abstract

Corn stover (stalks+leaves+cobs) is currently employed in the DOE-funded Biofuels Program as a model feedstock for integration of enzymatic cellulose hydrolysis-based biomass conversion technology. The chemical composition of corn stover is quite variable from lot to lot, and there are also indications that different lots of stover exhibit differential reactivity to dilute acid pretreatment, as practiced at NREL. These observations are also likely to apply to other herbaceous feedstocks. Biomass composition and quality have therefore emerged as important issues in the development of integrated conversion technology.

Since input feedstock chemical composition directly influences process yield, process economics can be dramatically impacted by changes in feedstock quality. Our goal is to determine the range of compositional variation as well as the potential causes of variability in mature corn stover. Over 1000 specimens from the 2001 North American crop were obtained from a wide variety of sources, including various commercial grain trials, experimental plots, and germplasm collection seed renewal plots. Samples therefore include a fairly broad cross-section of the maize germplasm, including commercial hybrids, inbreds, open-pollinated varieties, foreign accessions, primitive landraces, and related species. Samples were also obtained that had been produced under a variety of conditions in order to evaluate the relative extent to which genetic and environmental factors impact stover quality. We report here on the statistical analysis of the composition of over 700 stover specimens measured using a calibrated near-infrared spectroscopic method developed at NREL, including the results of an analysis of variance of these data as a function of genetic and environmental factors. Results show that corn stover composition varies over an unexpectedly large range and that both genetic and environmental factors are important influences on stover composition.

2. Methods

- Five to ten pound stover samples (10–20 stalks) were collected by suppliers at several institutions.
- Samples were thoroughly dried at 50ºC and milled individually to pass a 0.25-inch screen.
- Representative aliquots were selected for NIR spectroscopy.
- Compositions of samples were determined using a calibrated NIR/PLS1 method developed at NREL.
- Analysis of variance was performed using the Analysis ToolPak functions in Microsoft Excel 2001.



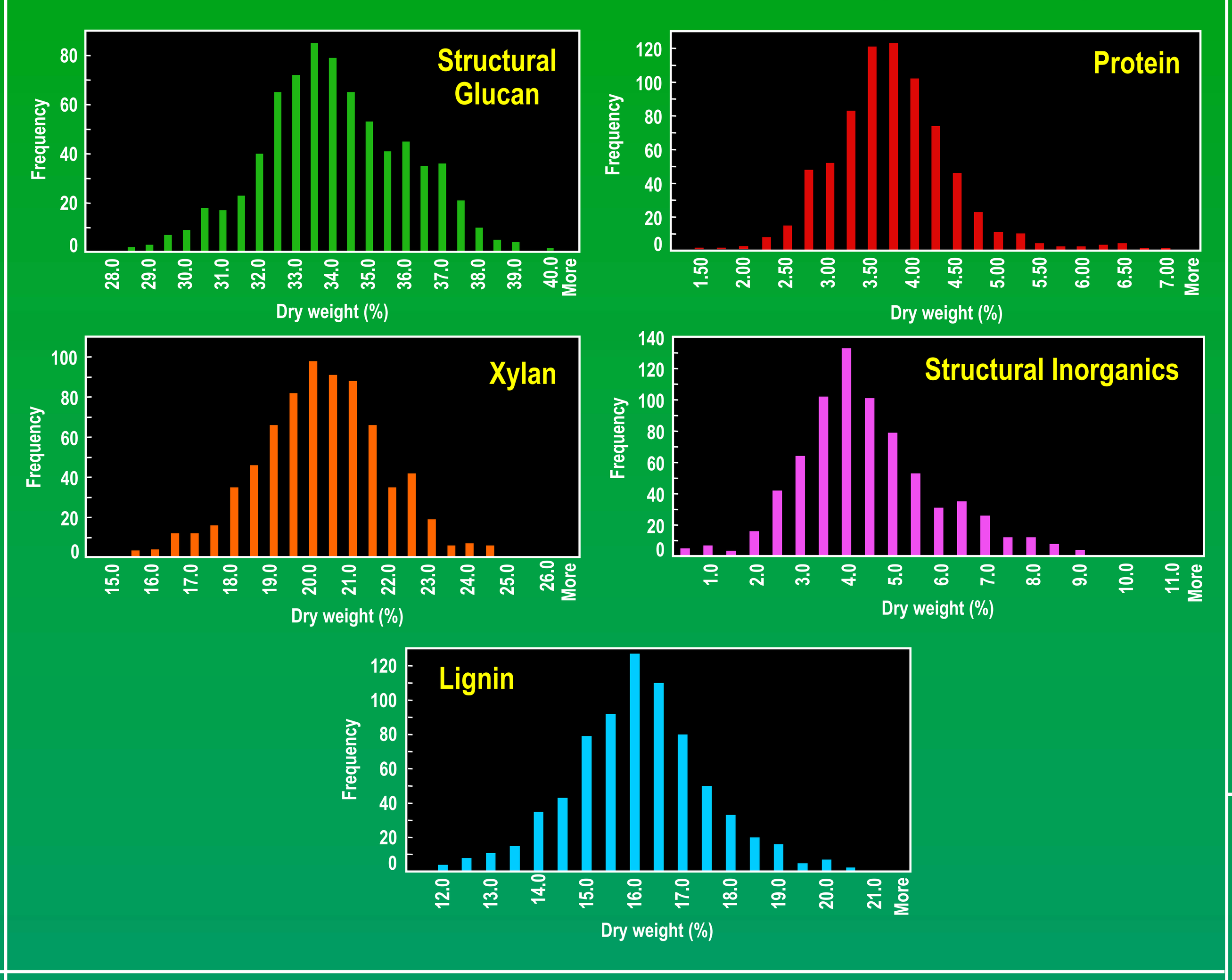
3. Sources of 2001 Corn Stover

Source	Contacts	# Samples	Comments
Monsanto	Diane Freeman, Brad Krohn, Dale Sorensen	221	this study
University of Minnesota	Dale Hicks, Tom Hoverstad, Steve Quiring	150	this study
University of Wisconsin, Madison	Joe Lauer	202	this study
USDA/ARS, Lincoln, NE	Wally Wilhelm	96	this study
University of Nebraska, Lincoln	Ken Russell	61	this study
Iowa State University	Wayne King	30	
ARS, Ames, IA	Linda Pollak	45	
ARS, NCPIS, Ames, IA	Mark Millard	242	
ProdiGene, Inc.	Donna Delaney, Beth Hood	14	
Total		1061	

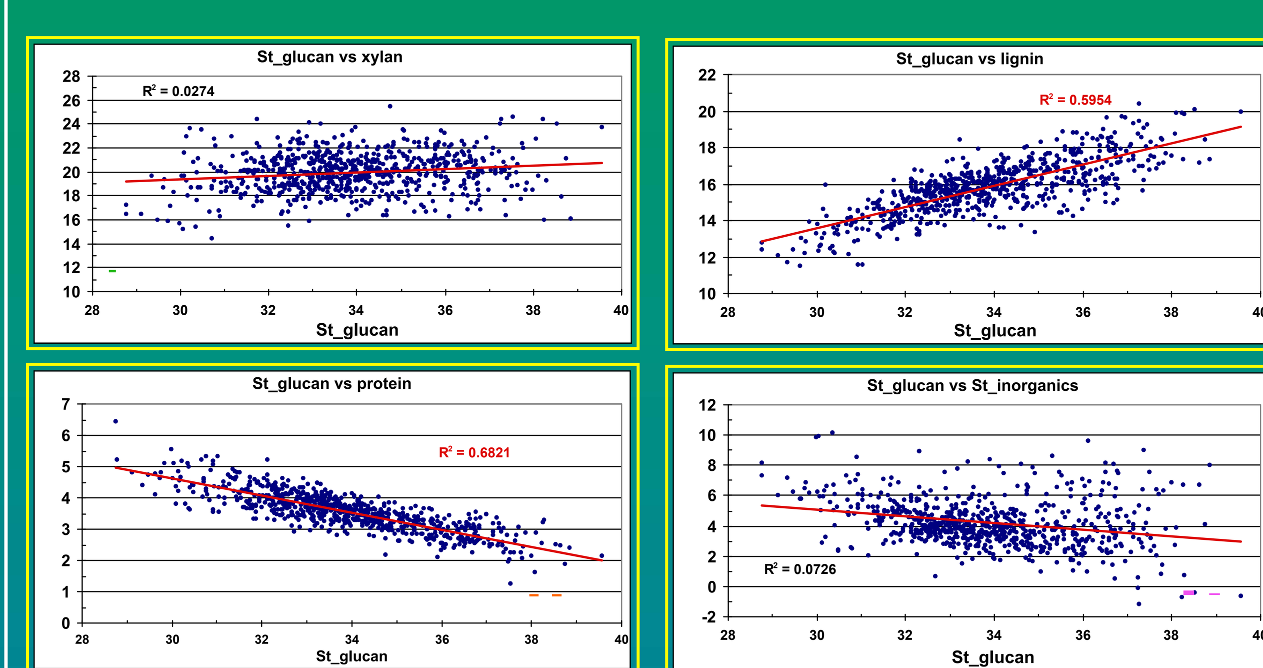
4. Population Statistics

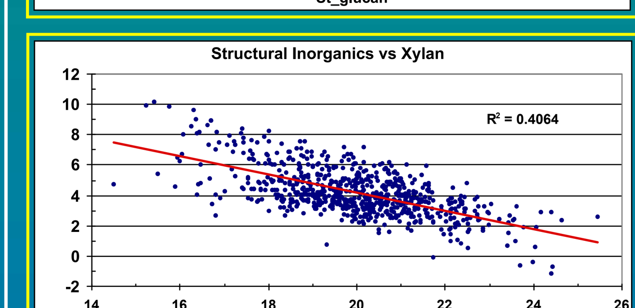
	Total_glucan	Struc_glucan	Xylan	Lignin	Protein	Acetyl	Uronic Acids	Structural Inorganics (Silica, Ash)	Soil	Calculated Soluble Solids	Total	Struc_glucan + Xylan	Total Structural Carbohydrates
Minimum (% dry wt.)	34.5	27.9	14.5	11.5	1.3	0.9	1.4	-1.2	0.9	2.0	90.0	43.3	45.3
Maximum (% dry wt.)	50.3	39.6	25.5	20.4	7.0	3.9	3.9	10.2	1.7	19.6	101.9	63.3	68.5
Span (% dry wt.)	15.8	11.7	11.0	8.9	5.7	3.0	2.5	11.3	0.8	17.5	11.9	19.0	23.2
Mean (% dry wt.)	42.0	33.8	20.0	15.8	3.6	2.7	2.9	4.2	1.3	8.2	97.4	53.8	58.7
Standard Deviation (% dry wt.)	1.5	2.0	1.6	1.4	0.7	0.5	0.3	1.6	0.1	2.2	1.7	2.8	3.2
Total Values	738	738	738	738	738	738	738	738	738	738	738	738	738

5. Population Distributions (n = 738)



6. Correlations Between Constituent Pairs



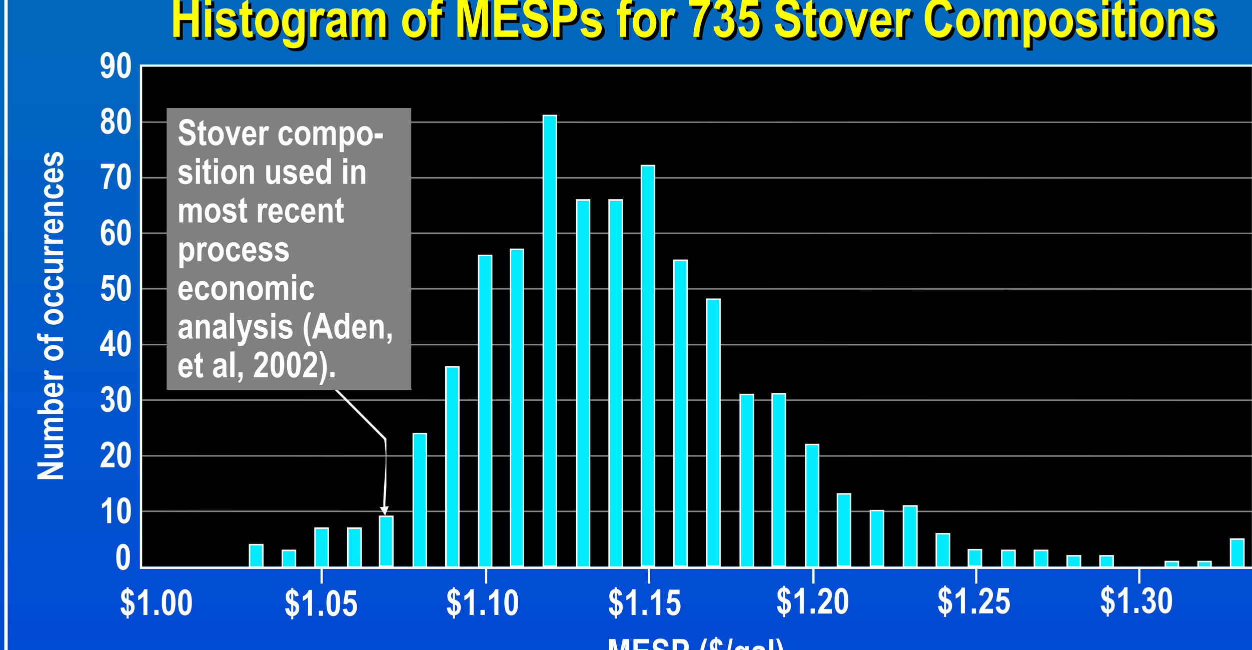


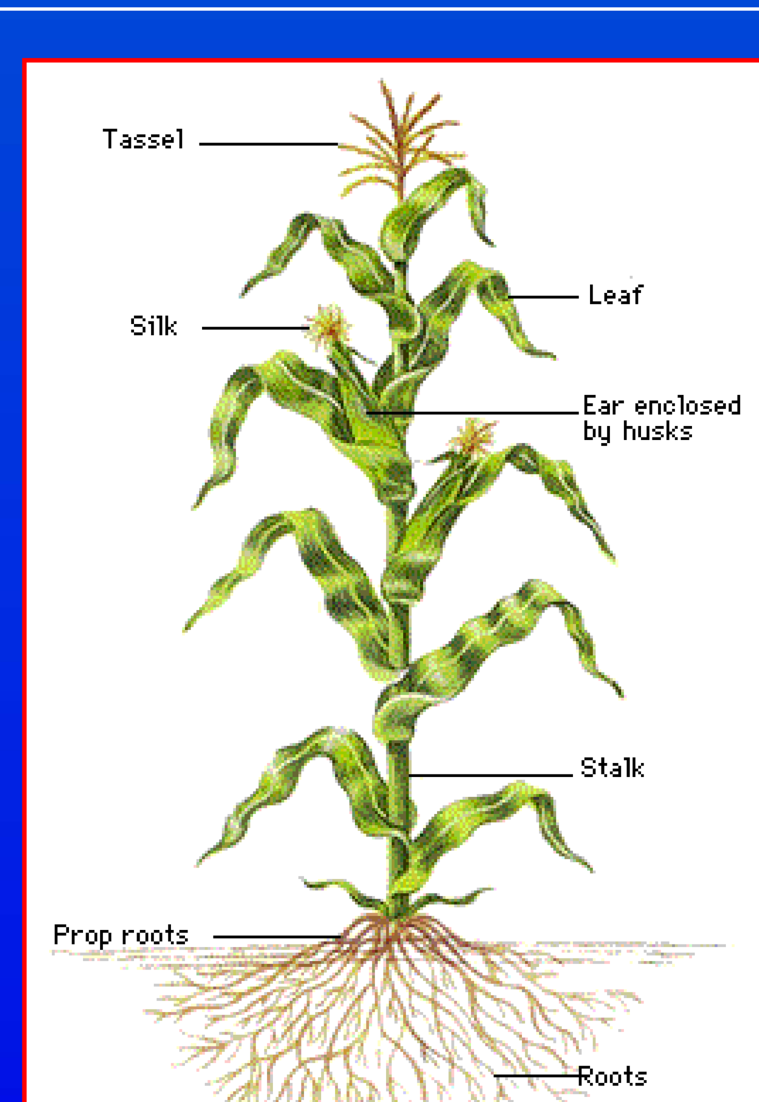
Correlation Coefficient Matrix

R^2	St_glucan	Xylan	Lignin	Protein	Acetyl	Str_inorg
Xylan	0.03					
Lignin	0.60	0.16				
Protein	0.70	0.18	0.32			
Acetyl	0.19	0.02	0.15	0.10		
St_inorg	0.10	0.41	0.32	0.13	0.12	
Soluble Solids	0.53	0.14	0.57	0.27	0.16	0.01

7. Effect of Stover Composition on Ethanol Cost

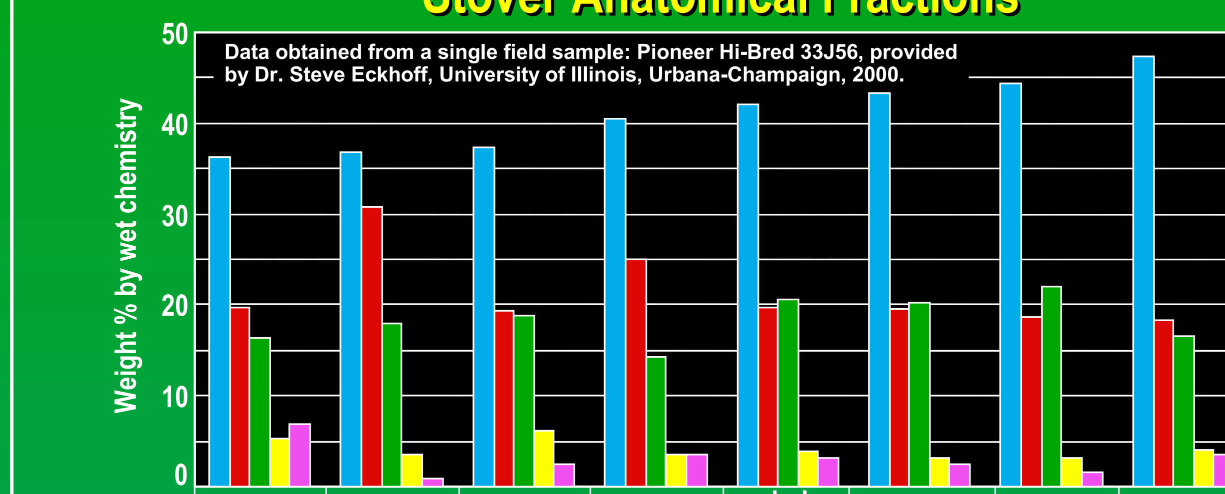
Histogram of MESPs for 735 Stover Compositions





8. Composition vs. Plant Anatomy

Stover Anatomical Fractions



	sheaths	cobs	nodes	leaves	whole stover	internodes	rind	pith
glucan	36.3	36.8	37.4	40.4	42.1	43.2	44.4	47.4
xylan	19.7	30.8	19.4	25.0	19.8	19.6	18.7	18.3
lignin	16.4	17.9	18.9	14.3	20.6	20.3	22.0	16.5
protein	5.2	3.6	6.2	3.5	3.8	3.1	3.2	4.0
inorganics	6.9	0.9	2.5	3.5	3.1	2.4	1.5	3.5

Fraction

9. ANOVA: University of Wisconsin Samples

Example: Structural glucan (South Central zone)

Raw data %Struc_glucan

Hybrid	Replicate	Galesville, WI	Fond du Lac, WI	Hancock, WI
Dekalb DKC51-88	1	32.76	33.66	32.57
	2	34.29	33.85	32.67
	3	32.22	34.33	33.39
Mycogen 4111	1	32.24	34.13	34.39
	2	33.36	34.00	33.59
	3	31.26	34.66	33.92
Northrup King N58-D1	1	32.44	35.77	33.09
	2	32.12	35.56	31.86
	3	31.77	35.10	31.93
Pioneer 37H26	1	32.14	35.24	32.18
	2	32.99	33.54	32.70
	3	31.30	35.12	33.16

ANOVA

ANOVA: Two-factor with replication — Struc_glucan

Source of Variation	SS	df	MS	F	P-value	F crit
Rows (genetics)	0.5644	3	0.18813	0.41495	0.7438103	3.00879
Columns (environment)	30.6548	2	15.3274	33.8064	1.04E-07	3.40283
Interaction (G x E)	9.92103	6	1.65351	3.64701	0.0102649	2.50819
Within	10.8813	24	0.45339			
Total	52.0215	35				

Summary of ANOVA results for major constituents across South Central Wisconsin

Summary of ANOVA results (P-values)

Constituent	Genetics	Environment	Interaction
Struc_glucan	0.744	1.04E-07	0.010
Xylan	0.015	4.30E-08	0.903
Lignin	2.02E-05	0.469	0.029
Protein	0.545	1.98E-08	0.135
St_inorg	0.008	4.97E-06	0.142
Struc_glucan + Xylan	0.181	8.38E-06	0.175

10. ANOVA: USDA/ARS, Lincoln Samples

Example: Xylan

Raw data %Xylan

Hybrid	Replicate	Non-irrigated No fertilizer	Non-irrigated fertilizer	Irrigated No fertilizer	Irrigated fertilizer
Pioneer 3162	1	16.31	17.42	20.01	19.60
	2	19.01	15.24	20.98	17.67
	3	17.90	21.22	20.69	20.27
Pioneer 3394	1	17.39	18.14	20.23	17.21
	2	18.63	19.89	17.62	17.55
	3	20.71	17.97	20.82	20.35
Pioneer 33R88	1	18.98	17.74	19.97	14.44
	2	18.21	18.99	16.88	14.93
	3	19.32	20.22	20.83	16.71
Pioneer 34G82	1	20.85	16.25	19.27	12.58
	2	17.13	18.75	19.54	13.33
	3	20.45	19.36	19.92	13.24
B73 x Mo17	1	20.66	19.72	18.47	17.82
	2	17.64	18.80	16.83	17.60
	3	18.87	19.16	18.55	17.19

ANOVA

ANOVA: Two-factor with replication — Xylan

Variation	SS	df	MS	F	P-value	F crit
Sample	14.8383	4	3.70957	1.79713	0.14845	2.60597
Columns	60.4766	3	20.1589	9.7661	5.8E-05	2.83875
Interaction	78.5386	12	6.54488	3.17072	0.00298	2.00346
Within	82.5666	40	2.06417			
Total	236.42	59				

Summary of ANOVA results for major constituents

Constituent	Genetics	Environment	Interaction
Struc_glucan	1.06E-07	1.28E-05	0.175
Xylan	0.148	5.81E-05	0.003
Lignin	0.002	0.062	0.112
Protein	1.46E-06	9.48E-11	0.435
St_Inorg	0.171	0.355	0.014
Struc_glucan + Xylan	0.001	0.002	0.086

11. Summary and Conclusions

- Corn stover composition varies widely
  - Total structural carbohydrates range from 45.3–68.5%
  - Statistically normal distributions for most major constituents
  - Stover composition has a large influence on Minimum Ethanol Selling Price (MESP)
  - The stover composition used to produce the most recent ethanol process economic model is overly optimistic
- A few weak correlations between constituent pairs
  - Struc_glucan positively with Lignin (r^2 = 0.60)
  - Struc_glucan inversely with Protein (r^2 = 0.69)
  - Xylan inversely with St_inorganics (r^2 = 0.47)
  - Struc_glucan inversely with Soluble Solids (r^2 = 0.56)
  - Lignin inversely with Soluble Solids (r^2 = 0.58)
  - All other pairs are not correlated
- Preliminary analysis of variance results indicate that stover composition varies as a function of:
  - Genetics
  - Location grown (environmental influences)
  - G x E interaction

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Charnell Clarke, summer intern (University of D.C.)

Jonathan Meuser, summer intern (UC Davis)