Penn State Dairy Cattle Nutrition Workshop

October 31 – November 1, 2018
Update on Lab Evaluations for Starch Digestibility and Other Feed Characteristics

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Cumberland Valley Analytical Services

CVAS’s new 33,000 sq. ft. facility devoted to feed and forage testing.
uNDF30 in Corn Silage Sept-YTD 2018 (CVAS data)

uNDF 30 (%DM) Distributions for Various Forage Types (CVAS, 2018)
Mycotoxin Survey in Corn Silage
2018 Crop, CVAS Data

<table>
<thead>
<tr>
<th>State</th>
<th>DON, ppm</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pennsylvania</td>
<td>2.1</td>
</tr>
<tr>
<td>New York</td>
<td>2.8</td>
</tr>
<tr>
<td>Ohio</td>
<td>2.2</td>
</tr>
<tr>
<td>Vermont</td>
<td>1.7</td>
</tr>
<tr>
<td>Wisconsin</td>
<td>3.1</td>
</tr>
<tr>
<td>Minnesota</td>
<td>2.7</td>
</tr>
</tbody>
</table>
Sampling

- Corn silage sample sent to the lab, perhaps 150 grams

- The difference of 10 corn kernals can move starch by 2 to 3 units ...
### Starch Evaluation by NIR

CVAS Calibration Statistics

<table>
<thead>
<tr>
<th></th>
<th>N</th>
<th>Mean</th>
<th>RSQ</th>
<th>SEC</th>
</tr>
</thead>
<tbody>
<tr>
<td>Corn Silage</td>
<td>1677</td>
<td>28.1 %</td>
<td>.98</td>
<td>1.01</td>
</tr>
<tr>
<td>Corn Grain</td>
<td>1302</td>
<td>71.2 %</td>
<td>.99</td>
<td>.45</td>
</tr>
</tbody>
</table>
Comparison of Starch by Chemistry and NIR

![Comparison graph]

$r^2 = .94$

$\text{RMSE} = 1.21$

Modeling Starch

- Modeling from aNDF
  - 2720 samples
  - $R^2: .73$
  - RMSE: 3.27
  - Equation:
    \[\text{Starch} = 80.4 - 1.17(\text{aNDF})\]
Modeling Starch Content in Corn Silage

- Modeling from CP, NDF, Ash
  - 2718 samples
  - $R^2$: .81
  - RMSE: 2.71
  - Equation:
    \[ \text{Starch} = 89.4 - 1.48(\text{CP}) - 1.03(\text{aNDF}) - .678(\text{ash}) \]

Modeling Starch Content in Corn Silage

- Modeling from CP, NDF, Soluble Fiber, Ash, Fat, Sugar, Lactic acid, Acetic acid
  - 40737 samples
  - $R^2$: .96
  - RMSE: 1.32
  - Equation:
    \[ \text{Starch} = 106.3 - .574(\text{CP}) - 1.10(\text{aNDF}) - 1.14(\text{soluble fiber}) - .978(\text{ash}) - 3.38(\text{fat}) - 1.28(\text{sugar}) - .567(\text{lactic}) - .108(\text{acetic}) \]
Relationships among in vitro starch digestibility (IVSD), rate of digestion, and predicted starch digestibilities (SD) in the rumen (kp = 0.089 h⁻¹) and total tract of dairy cows.

<table>
<thead>
<tr>
<th>IVSDa (% of starch)</th>
<th>Starch Kd (%)/h</th>
<th>Predicted Ruminal SDc</th>
<th>Predicted Total Tract SDd</th>
<th>Predicted Fecal Starch (%DM)e</th>
</tr>
</thead>
<tbody>
<tr>
<td>25f 4.8%</td>
<td>35.0%</td>
<td>88.7%</td>
<td>9.0</td>
<td></td>
</tr>
<tr>
<td>35 7.2%</td>
<td>44.7%</td>
<td>90.5%</td>
<td>7.6</td>
<td></td>
</tr>
<tr>
<td>45 10.0%</td>
<td>52.8%</td>
<td>92.0%</td>
<td>6.4</td>
<td></td>
</tr>
<tr>
<td>55 13.3%</td>
<td>59.9%</td>
<td>93.3%</td>
<td>5.4</td>
<td></td>
</tr>
<tr>
<td>65 17.5%</td>
<td>66.3%</td>
<td>94.5%</td>
<td>4.4</td>
<td></td>
</tr>
<tr>
<td>73 23.1%</td>
<td>72.2%</td>
<td>95.6%</td>
<td>3.5</td>
<td></td>
</tr>
<tr>
<td>85 31.6%</td>
<td>78.0%</td>
<td>96.7%</td>
<td>2.7</td>
<td></td>
</tr>
<tr>
<td>95 49.9%</td>
<td>84.9%</td>
<td>97.9%</td>
<td>1.7</td>
<td></td>
</tr>
<tr>
<td>99f 76.8%</td>
<td>89.6%</td>
<td>98.8%</td>
<td>1.0</td>
<td></td>
</tr>
</tbody>
</table>

a In vitro starch digestibility measured after 7 h of fermentation.
b Rate of starch digestibility (Kd) calculated from IVSD (7 h) using the method of Mertens Innovation & Research LLC for a single pool of starch with a lag time of 1 h and no indigestible starch (MIR_kd_P1T1u0TM).
c Ruminal starch digestibility calculated using the steady-state formula: Ruminal SD% = 100*(Kd / (Kd + kp)) assuming a kp for starch of 6.69%/h.
d Adjusted Total Tract Starch Digestibility = 82.224 + 0.185*Ruminal Starch digestibility –0.002. (Ferrareto et al., 2013)
e Total Tract Starch Digestibility = 100.0 – 1.25*Fecal starch (% fecal DM) solved for fecal starch. (Fredin et al., 2014)

These extreme values are rarely measured, but provide limiting boundaries on ruminal and total tract starch digestibilities when using MIR_kd.
Distribution of Starch kd (%/hr), 4mm grind, CVAS 2016 to 2018 for Corn Silage

The StarchD Challenge

• The industry says:

“The use of starch digestibility information from labs is not working.”

Translated: When I use the numbers from the lab they are not consistent with anticipated cow response.
The StarchD Challenge

- Limitations in the process:
  - StarchD is a complex and not fully understood process: What are we attempting to measure?

  Starch digestibility is not a “nutrient” - it is a complex set of interactions defined within a dynamic and complex rumen environment.

Impacting StarchD

- Particle size
- Fragility
- Moisture
- Weather impact during ear development
- Dry-down of kernels in the field
- Fermentation
- Time in fermentation
- Protein, Zein protein, Zein-starch ratio
- Amount of shell relative to starch flour
- Other physiological characteristics of starch in kernel
## Corn Grain Fragility – 4mm knife mill
(CVAS, 2014)

<table>
<thead>
<tr>
<th>Corn Type</th>
<th>MPS (microns)</th>
<th>Surface Area (cm² gm)</th>
<th>Particles / gm</th>
<th>IVSD7 (% Starch)</th>
<th>IVSD7 (% Starch) (1mm grind)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Floury</td>
<td>848</td>
<td>65.5</td>
<td>7788</td>
<td>73.7</td>
<td>83.5</td>
</tr>
<tr>
<td>Hybrid</td>
<td>905</td>
<td>61.5</td>
<td>6282</td>
<td>57.6</td>
<td>66.5</td>
</tr>
<tr>
<td>Flint</td>
<td>966</td>
<td>57.9</td>
<td>5632</td>
<td>50.6</td>
<td>61.9</td>
</tr>
</tbody>
</table>

### Cutter Mill for executing 4mm grind
Impacting StarchD

- Cow Perspective:
  - Particle size as fed
  - Mastication
  - Level of starch fed
  - Associative effects
  - Rumen environment
  - Time of feeding
  - Various enzyme concentrations
  - Rate of passage

Figure 4. Effect of grinding size on ruminal in situ starch disappearance (% of starch) at 0, 3, 6, 18, 24, 48, 72, 96 and 120 h of dry dent corn.
The StarchD Challenge

• Limitations in the process:
  – StarchD is a complex and not fully understood process: What are we attempting to measure?
  – Lab limitations require that our sample for analysis is not what we feed the cow.

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  – Lab limitations require that our sample for analysis is not what we feed the cow.
  – For routine use in the context of ration work we need to move evaluation process to NIR – this is problematic.
NIR Grind vs Analyzed Grind

NIR Grind vs Analyzed Grind
The StarchD Challenge

• Limitations in the process:
  – StarchD is a complex and not fully understood process: What are we attempting to measure?
  – Lab limitations require that our sample for analysis is not what we feed the cow.
  – For routine use in the context of ration work we need to move evaluation process to NIR – this is problematic.
  – The use of in vitro analysis for starch digestibility analysis has large inherent variability.

The StarchD Challenge

• Limitations in the process:
  – Lab data does not necessarily “fit” into current models.
  – Rate of passage significantly impacts realized rumen starch degradability and by implication how we want to benchmark StarchD.
The StarchD Challenge

• Limitations in the process:
  – Lab data does not necessarily “fit” into current models.
  – Rate of passage significantly impacts realized rumen starch degradability and by implication how we want to benchmark StarchD.
  – We have an incorrect mind-set as to use the information at hand.
The StarchD Challenge

• Productive mindset:

Consider StarchD (IVSD) information as a tool to rank feeds, to understand feed potential or limitations, and to bias nutritional models.

We need to realize that IVSD information at this time needs to be used more in a qualitative manner, not quantitative.

In Vitro Starch Digestibility, 7 hr, 4mm
(CVAS, chemistry, 2014 - 2017 crop years)

N= 3522
Ave. = 68.5
StDev = 9.52
Where do we go from here?

- Modeling approach to estimating static IVSD:
  - Particle Size
  - Moisture
  - Fermentation
  - Protein Fractions
  - Multiple time points to estimate rates

Effect of ensiling time on starch digestibility in HMC

Ferraretto et al., 2014
Modeling IVSD7 in Corn Grain

Summary of Fit

- **RSquare**: 0.766364
- **RSquare Adj**: 0.768323
- **Root Mean Square Error**: 2.40158
- **Mean of Response**: 61.38398
- **Observations (or Sum Weights)**: 465

**Effect Summary**

<table>
<thead>
<tr>
<th>Source</th>
<th>FDR LogWorth</th>
<th>FDR PValue</th>
</tr>
</thead>
<tbody>
<tr>
<td>CP</td>
<td>74.975</td>
<td>0.00000</td>
</tr>
<tr>
<td>ADF</td>
<td>14.975</td>
<td>0.00000</td>
</tr>
<tr>
<td>Acetic</td>
<td>10.691</td>
<td>0.00000</td>
</tr>
<tr>
<td>DM</td>
<td>4.945</td>
<td>0.00001</td>
</tr>
<tr>
<td>pH</td>
<td>4.798</td>
<td>0.00002</td>
</tr>
<tr>
<td>Lactic</td>
<td>1.129</td>
<td>0.07436</td>
</tr>
</tbody>
</table>

Modeling IVSD7 in Corn Grain

**Response IVSD 7hr, 4mm, %starch**

Effect Summary using CNCPS Protein Fractions

<table>
<thead>
<tr>
<th>Source</th>
<th>FDR LogWorth</th>
<th>FDR PValue</th>
</tr>
</thead>
<tbody>
<tr>
<td>A2, %DM</td>
<td>15.849</td>
<td>0.00000</td>
</tr>
<tr>
<td>B1, %DM</td>
<td>8.280</td>
<td>0.00000</td>
</tr>
<tr>
<td>B2, %DM</td>
<td>36.278</td>
<td>0.00000</td>
</tr>
<tr>
<td>A1, %DM</td>
<td>32.812</td>
<td>0.00000</td>
</tr>
<tr>
<td>DM</td>
<td>20.016</td>
<td>0.00000</td>
</tr>
<tr>
<td>pH</td>
<td>2.985</td>
<td>0.00103</td>
</tr>
</tbody>
</table>
Background

• The current corn silage processing score (CSPS) was designed to determine the proportion of starch in small particles (Mertens, 2005) that would most likely be fermented without additional chewing

  – 600 ml test sample is selected from undried silage after mixing – sampling undried material is more representative
  – Test sample is dried at <60°C to improve separation of starch from vegetative matter (hypothetically)
Background

- The current corn silage processing score (CSPS)

  - Dried sample is placed on a stack of ten sieves (20-cm diameter), with square apertures ranging from 19.0 to 0.30 mm, plus a bottom pan
  - Sample is vigorously shaken vertically using a Ro-Tap shaker for 10 min.
  - Sample retained on sieves with >4.75-mm apertures are combined and analyzed for starch
  - CSPS = 100 * (Total_Starch - >4.75-mm_Starch) / Total Starch

Rotap shaker showing 4.75mm screen and corn retained on the sieve
Distribution of Corn Silage Processing Scores, CVAS, 2015 and 2016 Crop Years

N= 5292
Ave. = 62.6
StDev = 11.4

Distribution of Corn Silage Processing Scores, CVAS, 2015 and 2016 Crop Years

Industry Makes Advances in Corn Silage Processing Past 12 YRS

<table>
<thead>
<tr>
<th>Crop Year</th>
<th>Number</th>
<th>Average</th>
<th>Percent Optimum</th>
<th>Percent Poor</th>
</tr>
</thead>
<tbody>
<tr>
<td>2006</td>
<td>97</td>
<td>52.8</td>
<td>8.2</td>
<td>43.3</td>
</tr>
<tr>
<td>2007</td>
<td>272</td>
<td>52.3</td>
<td>9.2</td>
<td>37.9</td>
</tr>
<tr>
<td>2008</td>
<td>250</td>
<td>54.6</td>
<td>5.2</td>
<td>34.8</td>
</tr>
<tr>
<td>2009</td>
<td>244</td>
<td>51.1</td>
<td>6.1</td>
<td>48.0</td>
</tr>
<tr>
<td>2010</td>
<td>373</td>
<td>51.4</td>
<td>5.9</td>
<td>43.4</td>
</tr>
<tr>
<td>2011</td>
<td>726</td>
<td>55.5</td>
<td>12.3</td>
<td>33.1</td>
</tr>
<tr>
<td>2012</td>
<td>871</td>
<td>60.8</td>
<td>14.8</td>
<td>19.9</td>
</tr>
<tr>
<td>2013</td>
<td>2658</td>
<td>64.6</td>
<td>26.2</td>
<td>22.1</td>
</tr>
<tr>
<td>2014</td>
<td>4634</td>
<td>62.2</td>
<td>25.8</td>
<td>10.4</td>
</tr>
<tr>
<td>2015</td>
<td>3231</td>
<td>61.1</td>
<td>24.2</td>
<td>17.5</td>
</tr>
<tr>
<td>2016</td>
<td>3598</td>
<td>63.5</td>
<td>30.8</td>
<td>11.5</td>
</tr>
<tr>
<td>2017</td>
<td>3349</td>
<td>65.4</td>
<td>38.2</td>
<td>9.4</td>
</tr>
</tbody>
</table>
Apparent (whole tract) Digestibility

• There has been interest in evaluating fecal starch as an indicator of digestion efficiency.
• This approach does not account for beginning starch level or the concentration effect in the manure.
• Several U.S. labs are using undigested NDF at 240 hr in vitro incubation (uNDF240) as a marker to relate the starting and ending starch levels.

Apparent (whole tract) Digestibility

• CVAS has developed NIR equations for 240 hour indigestible NDF in TMR and fecal material.
• Clients submit samples of TMR and associated fecal material to the laboratory.
• CVAS provides an analysis of the TMR and fecal material and a report of Apparent Digestibility for Starch and pdNDF.
• This information can be used as a diagnostic tool to evaluate ration efficiency and for educating the producer about nutritional concepts.
Background

• The current corn silage processing score (CSPS)
  -- Advantages
    • Provides a quantitative measure of kernel fragmentation
    • Particle size distribution of DM is determined simultaneously
    • Determining NDF on sieves with <1.18-mm apertures and the pan provides a simultaneous estimate of the peNDF of the silage
  -- Disadvantages
    • Only 4 to 5 samples can be analyzed per hour not including starch analysis time
    • Vertical shaking measures the width of particles and not length
    • Small starch particles may stick to large vegetative particles – particular concern for immature corn silages with soft starch
    • Drying may cause starch particles to adhere to each other
Developed Method

- Wet corn silage processing score (WCSPS)
  - Undried submitted material was mixed
  - 150g test sample of undried material was obtained
  - Test sample was placed in a 20.3 cm tall by 30.5 cm diameter sieve with 4.75 mm square apertures
  - Sample in sieve was plunged into a tank of room temperature water with a stroke of 10 cm (at the top of the stroke the sample was out of the water)
  - Sample was wet-sieved for 90 sec using a frequency of 60 strokes per min.
Developed Method

- Wet corn silage processing score (WCSPS)
  - After rinsing, residue retained by the sieve was dried, ground, and starch concentration was determined
  - WCSPS = 100 * (Total_Starch – Starch Retained) / Total Starch

Dry CSPS Relationship to DM

- Assuming similar processing, we might expect higher CSPS with lower DM, but the relationship is poor and slightly positive
- Perhaps drier corn silages are processed more extensively?
Proportion of DM retained on >4.75-mm sieves does not indicate more extensive processing of drier corn silages when dry sieved.

Wet sieving resulted in more DM and less starch retained on sieves with 4.75-mm apertures than did sieving dried corn silages.

This observation is consistent with the hypothesis that wet sieving removes starch that is not removed by sieving dried corn silages.

Wet sieving retains more DM, which is probably due to separation by flotation in which particles are sorted more by length, whereas dry separation using vigorous vertical shaking sorts by smallest dimension of the particles (width).
Relationships between Dry and Wet CSPS

The variation among samples was similar between sieving methods for retention of DM and starch.

The average CSPS of 189 corn silages was slightly higher when measured using the wet compared to the dry method.

The wet CSPS measured greater variation among corn silages than the dry method.

<table>
<thead>
<tr>
<th></th>
<th>DM &gt; 4.75 mm</th>
<th>Starch &gt;4.75 mm</th>
<th>CSPS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wet Retained</td>
<td>45.33</td>
<td>8.45</td>
<td>72.93</td>
</tr>
<tr>
<td>Dry Retained</td>
<td>38.20</td>
<td>9.15</td>
<td>70.49</td>
</tr>
<tr>
<td>Wet St Dev</td>
<td>7.30</td>
<td>3.66</td>
<td>11.03</td>
</tr>
<tr>
<td>Dry St Dev</td>
<td>7.25</td>
<td>2.93</td>
<td>9.06</td>
</tr>
</tbody>
</table>

DM > 4.75 mm retained

- DM retained on 4.75-mm sieves were moderately correlated between wet and dry methods.
- The slope of geometric equations were near 1.00 and the intercept indicates that wet sieving retentions were about 6.4% of DM higher than dry sieving.
Wet CSPS Relationship to CSPS

- Relationship is moderate suggesting that wet and dry CSPS measure slightly different things.
- When below 70, CSPS is higher than wet CSPS.

Wet CSPS Relationship to CSPS

- Although the R² is low, there is a trend with wet CSPS generating higher differences when CSiL DM is <35%.
- Dry CSPS generates lower differences when CSiL DM is >35%.
Differences in CSPS and Wet CSPS by Corn Silage DM Ranges

<table>
<thead>
<tr>
<th>DM range</th>
<th>Percent of samples</th>
<th>Avg DM</th>
<th>Avg CSPS</th>
<th>Avg Wet CSPS</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
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<td></td>
<td></td>
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</tbody>
</table>

For corn silages < 35% DM, wet CSPS were higher than CSPS suggesting that the wet method removes more starch than the dry method.

Distribution of Fecal Starch in Dairy TMR (CVAS, 2017)

- N = 1576
- Ave. = 4.11
- StDev = 2.30

![Graph showing distribution of fecal starch in dairy TMR](image)
Fecal starch interpretation

- < 3% fecal starch = good, no need to investigate individual feeds
- 5% fecal starch = Total tract starch digestibility ~93.75%. Potential to investigate individual feeds
- > 5% fecal starch = evaluate individual feeds and/or management practices

Adapted from Dr. Larry Chase

Calculated Rumen Starch Digestibility using Paired TMR and Fecal Samples (CVAS 2017)

N= 1576
Ave. = 64.2
StDev = 18.93
Penn State Dairy Cattle Nutrition Workshop

October 31 – November 1, 2018
Update on Lab Evaluations for Starch Digestibility and Other Feed Characteristics

Ralph Ward
Cumberland Valley Analytical Services