Practical Options for PEDv Mitigation During Feed Manufacturing

Steve Dritz
Cassie Jones
Jason Woodworth
Kansas State University

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K-State Swine Day
Prevention of Biological Hazards in Feed

• Why is PEDv mitigation important?
  – Control animal food safety hazards to fulfill our role in preserving farm-to-fork food safety.
K-State Study 1: PEDv is highly infectious!

- With PEDv, a dose as low as 200 infectious particles in feed has been demonstrated to result in pig infection.
- An acutely infected piglet can produce 100,000,000 infectious particles per gram of feces
- Thus, 1 gram of feces from an acutely infected pig could contaminate up to 500 tons of feed with each gram of feed being infectious
Cargill Feed Safety Research Center

• 3 story BSL-2 Lab
  – *Salmonella*, E. Faecium, PEDV
  – Pellet mills, coolers, and bagging capacities
  – Containment mode
    • Equipped with sanitation features
    • Air flow alarms
    • HEPA filters
    • Decontamination
KSU Study 2: What happens when PEDv contaminated feed is produced?

- 3 replications (days) of PEDv-inoculated feed mixed, discharged through a bucket elevator in the FSRC.
- Environmental swabs collected of equipment and facility surfaces after each batch and analyzed via PCR for detection of PEDv.
Environmental contamination after processing PEDv-inoculated feed

Zone 1 = direct feed contact surfaces - equipment interiors
Zone 2 = surfaces directly adjacent to zone 1
Zone 3 = structural surfaces - floors, walls
Preventing cross contamination during production: Housekeeping
Mitigation of Hazards

• Dust Collection

DO NOT add back to the feed!
KSU Study 3: Can PEDv Infectivity Be Reduced by Flushing or Sequencing Diets?

- Sequencing or flushing are recognized cleanout procedures for CGMPs of medicated feed
- One batch of PEDv-negative feed mixed, conveyed through bucket elevator, discharged
- One batch of PEDv-positive feed followed
- Four subsequent sequences of PEDv-negative feed followed
  - Feed and environmental samples at multiple locations collected after each batch
KSU Study 3: Can PEDv Infectivity Be Reduced by Flushing or Sequencing Diets?

Number of Feed Samples When PEDv was Detected by PCR

<table>
<thead>
<tr>
<th>Location</th>
<th>Time Point</th>
<th>After PEDV Diet</th>
<th>After Sequence 1</th>
<th>After Sequence 2</th>
<th>After Sequence 3</th>
<th>After Sequence 4</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mixer</td>
<td>9/9</td>
<td>7/9</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Bucket Elevator</td>
<td>9/9</td>
<td>7/9</td>
<td>2/9</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

Number of Pigs Infected with PEDv by Bioassay

<table>
<thead>
<tr>
<th>Location</th>
<th>Time Point</th>
<th>After PEDV Diet</th>
<th>After Sequence 1</th>
<th>After Sequence 2</th>
<th>After Sequence 3</th>
<th>After Sequence 4</th>
</tr>
</thead>
<tbody>
<tr>
<td>2 dpi (fecal)</td>
<td>9/9</td>
<td>1/9</td>
<td>1/9</td>
<td>?</td>
<td>?</td>
<td>?</td>
</tr>
</tbody>
</table>
Feed mills decontamination can be a challenge.
KSU Study 4: Can Pelleting Reduce the Infectivity of PEDv?

- Low dose and high dose (20 and 13 Ct)
- 3 pellet mill conditioner retention times (45, 90, 180 s)
- 3 conditioning temperatures (155, 175, 195°F)

<table>
<thead>
<tr>
<th>Temp, °F</th>
<th>45</th>
<th>90</th>
<th>180</th>
</tr>
</thead>
<tbody>
<tr>
<td>155</td>
<td>43</td>
<td>40</td>
<td>45</td>
</tr>
<tr>
<td>175</td>
<td>37</td>
<td>40</td>
<td>42</td>
</tr>
<tr>
<td>195</td>
<td>40</td>
<td>37</td>
<td>36</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Temp, °F</th>
<th>45</th>
<th>90</th>
<th>180</th>
</tr>
</thead>
<tbody>
<tr>
<td>155</td>
<td>30</td>
<td>30</td>
<td>30</td>
</tr>
<tr>
<td>175</td>
<td>30</td>
<td>30</td>
<td>30</td>
</tr>
<tr>
<td>195</td>
<td>30</td>
<td>31</td>
<td>30</td>
</tr>
</tbody>
</table>

Low Dose Feed No processing = 31
High Dose Feed No processing = 24

No infectivity developed from any of the pelleted diets
KSU Study 5: Is There Risk for PEDv Infectivity at Lower Conditioning Temperatures?

- Single dose (11 Ct)
- Single conditioner retention time (30 s)
- 5 conditioning temperatures (100, 115, 130, 145, and 160°F)
- Replicated manufacturing conditions
  - 3 pigs/room – one from each manufacturing rep

### PCR Ct Values

<table>
<thead>
<tr>
<th>Temp, °F</th>
<th>Time, sec</th>
</tr>
</thead>
<tbody>
<tr>
<td>100</td>
<td>32.5</td>
</tr>
<tr>
<td>115</td>
<td>34.7</td>
</tr>
<tr>
<td>130</td>
<td>37.0</td>
</tr>
<tr>
<td>145</td>
<td>36.5</td>
</tr>
<tr>
<td>160</td>
<td>36.7</td>
</tr>
</tbody>
</table>
**KSU Study 5: Is There Risk for PEDv Infectivity at Lower Conditioning Temperatures?**

Number of Pigs Infected with PEDv by Bioassay

<table>
<thead>
<tr>
<th>Feed</th>
<th>0 dpi</th>
<th>2 dpi</th>
<th>4 dpi</th>
<th>6 dpi</th>
<th>7 dpi</th>
<th>7 dpi Cecum</th>
</tr>
</thead>
<tbody>
<tr>
<td>No PEDV</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>100°F</td>
<td>9/9</td>
<td>0</td>
<td>1/9</td>
<td>3/9</td>
<td>3/9</td>
<td>3/9</td>
</tr>
<tr>
<td>130°F</td>
<td>9/9</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>145°F</td>
<td>8/9</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>160°F</td>
<td>8/9</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

Infectivity developed in diets pelleted below 130°F
KSU Study 5: Is There Risk for PEDv Infectivity at Lower Conditioning Temperatures?

• Thermal mitigation of PEDv by pelleting
  – When is feed NOT conditioned to at least 130°F?
    • Intentional extremely low conditioning temperatures (rare)
    • Start-up
    • Plugged dies
  – Other potential mitigation strategies may be necessary to consider IN ADDITION to pelleting
KSU Study 6: Can Chemicals be Added to Feed or Ingredients to Reduce the Risk of PEDv Cross-Contamination?

• 4 different feed or ingredients to be treated:
  – Complete nursery feed, porcine MBM, blood meal, SDAP

• 5 chemical treatments:
  – Organic acids, essential oils, sodium bisulfate, Termin-8, and sodium chlorate

• After the feed or ingredients were treated they were inoculated with PEDv (5.6 x 10^4 TCID 50/g)

• Samples evaluated on days 0, 1, 3, 7, 14, 21, and 42 after inoculation for determination of PEDv RNA via RT-qPCR
Untreated Controls stored at Room Temperature

Quantitative Ct Values

- Swine Diet
- Blood Meal
- Porcine Meat and Bone Meal
- Spray Dried Animal Plasma

Day

0 10 20 30 40 50

Quantitative Ct Values

24 26 28 30 32 34 36 38 40

K-State Research and Extension
Pork Checkoff
PEDv contamination post-treatment in swine diets stored at room temperature

- Untreated control
- Medium chain fatty acid
- Organic acid
- Essential oil
- Sodium bisulfate
- Sodium chlorate
- Commercial formaldehyde

Quantitative CT Values

Ingredient \times day, P < 0.0001
Treatment \times day, P < 0.0001
Ingredient \times treatment, P < 0.0001
KSU Study 7: What is the Role of MCFA on Preventing PEDv Infectivity in Various Ingredients

• 15 different ingredients or feed treated:
• 3 chemical treatments:
  – Control, SalCurb, 2% MCFA blend
• Samples evaluated on days 1, 8, 27, and 37
• All samples treated with MCFA and SalCurb were non-infectious.
KSU Study 8: What Quantity of MCFA is Needed to Prevent Infectivity of PEDv?

- Bioassay scheduled in December

[Graph showing comparison of different feed samples including Neg Feed, Positive Feed, Formaldehyde Feed, MCFA1 Feed, and MCFA2 Feed, with data for Neg Plasma and Positive Plasma depicted.]

- Main effects $P < 0.0001$
- Treatment $\times$ form $P < 0.0001$
- Treatment $\times$ day $P < 0.0001$
- Treatment $\times$ day $\times$ form $P < 0.02$

Data points for Feed Samples and SDAP Samples are indicated with different letters.
Summary of PEDV Findings

• Feed mill biosecurity is important to prevent cross-contamination of PEDv.

• Sequencing diets dilutes PEDv, but infectivity remains.
  – Particularly on equipment surfaces

• Diets pelleted with a 30 s conditioning time above 130°F were non-infectious.
  – Considerations required for plugs, start-up of pellet mills.

• Formaldehyde and MCFA demonstrate some ability to reduce PEDv RNA, but is ingredient dependent.
  – Effectiveness against infectivity and feasible concentrations in process of being determined.

• Multiple preventive and proactive strategies may need to be employed to maximize PEDv control.
Partners for our PEDv Research

• KSU Applied Swine Nutrition Team
  – Drs. Nitikanchana, Dritz, Woodworth, Tokach, DeRouchey, Goodband, Schumacher, Jordan Gebhardt

• KSU Grain Science
  – Drs. Jones, Huss, and Stark; Roger Cochrane

• KSU VDL
  – Drs. Hesse, Bai, Haus, Anderson, and their team

• ISU VDL
  – Drs. Main, Zhang, Gauger, and their team

• National Pork Board and USDA
Impact of Feed Processing on Pig Performance

Kansas State University
Applied Swine Nutrition Team
&
Grain Science and Industry
Evaluating pellet and meal feeding regimens on finishing pig growth performance and stomach morphology
Effects of pelleting regimen on ADG of 70 to 300 lb pigs

De Jong et al., 2015

2,100 pigs
118 days

abc P > 0.10
SEM = 0.03
Effects of pelleting regimen on F/G

De Jong et al., 2015
Effects of pelleting regimen on stomach morphology (combined ulceration & keratinization)

De Jong et al., 2015

abc \( P < 0.05 \)
SEM = 0.613
Effects of pelleting regimen on IOFC

De Jong et al., 2015

P > 0.10
SEM = 2.45
Effects of Grinding Corn through a 2-, 3-, or 4-High Roller Mill on Milling Characteristics and Pig Performance
Influence of roller mill configuration on F/G of 25-50 lb pigs

ADG: $P < 0.479$
F/G: $P < 0.122$

<table>
<thead>
<tr>
<th>Rolls</th>
<th>2</th>
<th>3</th>
<th>4-Fine</th>
<th>4-Course</th>
</tr>
</thead>
<tbody>
<tr>
<td>Particle Size</td>
<td>525</td>
<td>394</td>
<td>267</td>
<td>403</td>
</tr>
<tr>
<td>Std. Dev.</td>
<td>3.14</td>
<td>2.73</td>
<td>2.57</td>
<td>2.81</td>
</tr>
</tbody>
</table>

Gebhardt et al., 2015
Corn particle size affects feed preference of nursery pigs

**Particle size (µm) of corn is noted above columns**

\[ P = 0.01 \quad P = 0.89 \quad P = 0.01 \quad P = 0.01 \quad P = 0.01 \]

<table>
<thead>
<tr>
<th>Trial 1</th>
<th>Trial 2</th>
<th>Trial 3</th>
<th>Trial 4</th>
<th>Trial 5</th>
</tr>
</thead>
<tbody>
<tr>
<td>525</td>
<td>403 525</td>
<td>403</td>
<td>700</td>
<td>700</td>
</tr>
<tr>
<td>267</td>
<td>267</td>
<td>400</td>
<td>250</td>
<td></td>
</tr>
</tbody>
</table>

De Jong et al., 2013; Gebhardt, 2015; Bokelman, 2015
Influence of roller mill configuration on ADG of 88-287 lb pigs

Gebhardt et al., 2015
Influence of roller mill configuration on ADFI of 88-287 lb pigs

Gebhardt et al., 2015
Influence of roller mill configuration on F/G of 88-287 lb pigs

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<tr>
<th>Rolls</th>
<th>Particle Size</th>
<th>Std. Dev.</th>
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</thead>
<tbody>
<tr>
<td>2</td>
<td>572</td>
<td>3.02</td>
</tr>
<tr>
<td>3</td>
<td>484</td>
<td>2.94</td>
</tr>
<tr>
<td>4-Fine</td>
<td>295</td>
<td>2.55</td>
</tr>
<tr>
<td>4-Course</td>
<td>382</td>
<td>2.95</td>
</tr>
</tbody>
</table>

P = 0.147

Gebhardt et al., 2015
Influence of roller mill configuration on mill performance of 88-287 lb pigs

Grinding rate, ton/hr

<table>
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<th>4-Course</th>
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<tr>
<td>Std. Dev.</td>
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<td>2.94</td>
<td>2.55</td>
<td>2.95</td>
</tr>
</tbody>
</table>

Electricity cost, $/hr

Gebhardt et al., 2015
Influence of roller mill configuration on IOFC of 88-287 lb pigs

Gebhardt et al., 2015
Example sample of corn analyzed with or without flow agent

Kalivoda et al., 2015
What is the influence of shake time, sieve agitators, or flow agent?

- **Time, min:**
  - 10
  - 10
  - 15
  - 15
  - 15

- **Agitator:**
  - Yes
  - Yes
  - No
  - Yes
  - Yes

- **Flow agent:**
  - No
  - Yes
  - No
  - No
  - Yes

**µm**

- **Std Dev**
  - 2.23
  - 2.62
  - 2.09
  - 2.27
  - 2.63

*(a,b,c) P < 0.05*

*Kalivoda et al., 2015*
K-State Particle Size Procedures

• We will change our procedures to determine particle size:
  – Include flow agent (0.5 g fumed silica)
  – Same sieves and agitators
  – Same 10 minute shake time

• Results will have a lower mean particle size and a higher standard deviation
Summary of Feed Processing Research

• Pelleting improved G:F at expense of ulcers, removals
  – Rotating provided intermediate G:F, fewer removals than pellets alone
• Little benefit to fine grinding in nursery pig diets
• Fine grinding in finishing no benefit F/G or IOFC
• Flow agent improves particle size analysis
  – Future K-State results will be conducted using flow agent
  – Lower mean particle size and higher standard deviation