Development and comparison of a novel rapid sampling method to serum and tonsil scraping to detect PRRSV in acutely infected sows

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**Education:**
- 2021-: MSc/PhD student in VDPAM, ISU, US.
- 2010-2013 – MSc in PVM, CAU, China.
- 2016-2010 – DVM(equivalent). BYAU, China.

**Professional Experiences:**
- 2020–2022, **ASFV Research Scientist** in Swine Research Institute (SRI) of Newhope Co., ltd, Qingdao, China;
- 2013-2019, Swine Vaccine Technical Services Specialist / Manager/ Sales Director at China Animal Husbandry Industrial Co., ltd, China.
• Professional Experiences:

• **Interpreter** of Leman China Swine Conference (2017-2021, China);
• Interpreter of IPVS (2016, Dublin; 2018, Chongqing, China);
• Interpreter of OIE CSFV Reference Laboratory Technical Conference (2017, 2019, Beijing, China) ;
• **Professional Experiences:**

• Interpreter of FLY-V Seminar Series for Future Leaders of Young Veterinarians (2017, 2018, 2019, Henan, China);

• CAHIC-MSD AH JV project Coordinator and Interpreter (2014-2017).
Outline

• Introduction
  • Difference of elimination strategy between PRRSV(US) and ASFV (CN)

• Proof of concept for Tonsil oral scraping (TOSc) design and development
  • TOSc collector design
  • TOSc sample collection
  • TOSc sample processing
  • TOSc sample composition

• Comparison of detection rate and Ct values between TOSc and tonsil scraping (TSc) and serum in acutely infected sows
Introduction
Introduction

• Difference between PRRSV and ASFV elimination

  • PRRSV: Herd closure to rely on sow immunity to clear the virus based on SIR model;

  • ASFV: low contagiousness, without known/proven immunity, test-removal or (partial) depopulation based on early detection in SOWs
Load-Close-Homogenize for PRRSV

Hallmark: Consecutive PRRSV negative weaned piglets!

Gestation sows IA

Gestation sows IB

Gestation sows II

= Previously exposed and shedding

= Previously exposed, no or “unknown” shedding

= Not previously exposed, no shedding

Adapted from slides of Dr. Holtkamp's courtesy
Test removal for ASFV

Hallmark: Consecutive negative sows
Early detection at low prevalence in sows!

= Previously exposed and shedding
= Previously exposed, no or “unknown” shedding
= Not previously exposed, no shedding

Adapted from slides of Dr. Holtkamp’s courtesy
Proof of concept for TOSc-sampling tool for PRRSV

1. Delayed time-to-stability (TTS) calls for better understanding PRRSV ecology in sow populations following outbreaks.

2. No easy sample type for sows
   - Serum, tonsil scraping for sows:
     - Labor intensive,
     - Animal invasive,
   - Oral fluid:
     - Various success rate

Objective: an easy and reliable sow samples
Without snaring!

The missing link of easy sampling in gestation sows
Proof of Concept for TOSc Design and Development
Tonsil Scraping for finisher and sows

- Restraining the sows/finishers
- Mucous like fluid

source: SMEC, Clinical Skills: Tonsil Scrapings (iastate.edu)
Tonsil-Oral Scraping method (TOSc)

**TOSc collector**-design of proof of concept

Rubber finger pad (thimble) to make it “ABRASIVE” to mimic tonsil scraping
TOSc: Sample Collection Process

Before sampling | Sample collection (w/o restraining) | Sample collected | TOSc fluid in tubes
Key points of TOSc collection process:

- In parallel with the vertical axis of mouth; (move)
- Depth with 5 inches of handle outside the mouth
- Upwards angle
- No resistance when scraping (hard palatine or bitten by the sow)
- Helpful when some sow is pushed frontwards especially for gilts.
TOSc – Sample Processing

Vortex & transfer

supernatant →

deposit →
What’s TOSc sample?

Figure 2. Sample 1. Higher magnification of figure 1. Focally on the left there is a small cluster of immune cells including lymphocytes, plasma cells, suspect macrophages, and rare eosinophils. Frequent squamous epithelial cells are also present.

Large amount of squamous cells and **some immune cells**
Comparison of detection rate and Ct values between TOSc and tonsil scraping (TSc) and serum in acutely infected sows
Study design

• Recent outbreak of PRRSV on a previously naïve sow farm
• 30 sows in farrowing
Comparison of sampling types for sows in terms PRRSV PCR positivity, Ct values, and need for straining.

<table>
<thead>
<tr>
<th>Sample type</th>
<th>Serum</th>
<th>Tonsil Scraping</th>
<th>Tonsil Oral scraping (TOSc)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Detection rate [95% CI]</td>
<td>16.8%&lt;sup&gt;a&lt;/sup&gt; [7.1%,34.3%] (5/30)</td>
<td>73.3%&lt;sup&gt;b&lt;/sup&gt; [60.2%,85.3%] (22/30)</td>
<td>100%&lt;sup&gt;ab&lt;/sup&gt; [87.7,99.9%] (30/30)</td>
</tr>
<tr>
<td>Average and range of PCR Ct values</td>
<td>35.2 [33.6-36.4]</td>
<td>30.7 [25.1-35.4]</td>
<td>29.7 [24.1-36.3]</td>
</tr>
<tr>
<td>Need for straining sows</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
</tr>
</tbody>
</table>

<sup>ab</sup>: different superscript letters indicate significant difference in least square means (Tukey test, p<0.05)
Ct ≤ 37 was deemed positive.

TOSc: Tonsil Oral Scraping;

***, p < 0.001; **, p < 0.01;

ns: not significant, p > 0.05 (Dunn Test).
Conclusion

• TOSc recovered a mixture of oral fluids and tonsil exudates within seconds without restraining the sows.

• Numerically, TOSc samples had higher PRRSV qPCR detection rate compared to serum and tonsil scraping and lower average Ct values than tonsil scraping and serum in acutely infected sows.

• Statistically, In terms of PRRSV RNA detection rate and Ct values, there was no significant difference between TOSc and tonsil scraping; while there was a significant pairwise difference between serum and tonsil scraping, and between TOSc and serum.

• Great potential of TOSc as a novel, practical, and rapid tool for PRRSV RNA detection in sows to be able to assess sow herd status.
Limitations

• Limited sample size

• Early infection status

• No comparison between TOSc and OF

• Further research needed for comparison between TOSc, OF, serum, tonsil scraping at different stages of infection with a larger sample size.
Thank you for guiding me!!

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