Disinfection Effectiveness against PRRSV and TGEV under Trailer Conditions

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Background and Justification
The modern swine industry is structured such that frequent movements of pigs are necessary. Transportation events pose a risk to swine for transmission of undesirable pathogens. Porcine reproductive and respiratory syndrome virus (PRRSV), in the family Arteriviridae, and transmissible gastroenteritis virus (TGEV), in the family Coronaviridae, are two viruses with large economic consequences that are capable of being transmitted by transport vehicles. Sanitation procedures to decrease the pathogen load in standard equipment used in swine transportation have been described, but trailers, sorting equipment, and chutes are not always thoroughly cleaned between every transport event and can pose a risk to negative herds. The purpose of this research was to compare Accel, an accelerated hydrogen peroxide disinfectant, (Virox Technologies Inc., Oakville, Ontario, Canada) to Synergize, a frequently used quaternary ammonium compound and glutaraldehyde combination disinfectant (Preserve International; Reno, Nevada) against PRRSV and TGEV in simulated field conditions using model trailers. The study was designed to enable replication of features, such as pooling of water on the floor and scattered pieces of fecal material throughout the model trailer, that are commonly encountered in full-sized trailers used for live-haul or market pigs. The first objective assessed the effectiveness of wash and disinfection protocols using Accel and Synergize with commonly used washing methods and relatively short disinfectant contact times against PRRSV and TGEV. The second used a bioassay to determine whether or not a sufficient quantity of live PRRSV and TGEV were present in the model trailers to infect pigs when inoculated intramuscularly (IM).

Materials and Methods

Objective 1
The experimental design for Objective 1 was an incomplete block design using three experimental units per block. The experimental unit in each block of this study was a model trailer measuring 0.62 meter (m) wide x 0.82 m tall x 1.11 m long. Four treatment groups were tested. Treatment 1 used Accel disinfectant and Treatment 2 tested Synergize disinfectant. Treatment 3 served as a positive control group and Treatment 4 was a negative control group.
Model trailers for Treatments 1, 2, and 3 were contaminated with PRRSV and TGEV. Two milliliters (ml) of each virus was mixed with 46 ml of a 1:1 deionized water/confirmed PRRSV and TGEV negative feces mixture and spread over the interior of the model trailer. Model trailers used in Treatment 4 were soiled using the uncontaminated water/feces mixture. All trailers were decontaminated using a high-pressure wash for 90 seconds with cold water; this cleaning method resulted in visible areas of puddling, scattered pieces of fecal material, and a few sections of the trailer walls and floor that were untouched by the power-washing procedure. Treatments 1 and 2 were disinfected using Accel and Synergize disinfectant, respectively. Treatment 3 was sham-disinfected using tap water. Treatment 4 was not disinfected.

Four samples at 5 time points were collected for each trailer replicate. Time points included pre-wash, immediately post-wash, and at 15, 30, and 60 minutes post-disinfection (Treatments 1 and 2; sham-disinfection for Treatment 3) or post-wash (Treatment 4). For each replicate, all 4 samples from each time point were pooled and tested for the presence of PRRSV ribonucleic acid (RNA) by polymerase chain reaction (PCR) and quantitative PCR (qPCR). The same pooled samples were tested for TGEV RNA by PCR at the Iowa State University Veterinary Diagnostic Lab.

**Objective 2**
A bioassay was completed for Objective 2. Twenty-four 10-week-old PRRSV and TGEV negative pigs were assigned to one of six bioassay groups to test whether or not infectious live virus was present in supernatant from samples collected for Objective 1. Table 1 identifies the bioassay groups by time points tested. All samples from the Objective 1 treatment groups were pooled by time point to prepare the inoculum. Pigs in Bioassay Groups 1 to 5 were challenged with a minimum of 4 ml of supernatant by IM injection and 7 ml by oral gavage. Bioassay groups were housed by group and monitored for 14 days. Blood and fecal samples were collected at 7 days and 14 days post-inoculation (PI) and tested for the presence of PRRSV RNA or TGEV RNA by PCR at days 7 and 14 PI and for the presence of PRRSV or TGEV antibodies by enzyme-linked immunosorbent assay (ELISA) on day 14 PI. Transmissible gastroenteritis virus immunohistochemistry (IHC) was completed on harvested intestinal tissue at day 14.

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<thead>
<tr>
<th>Bioassay Group</th>
<th>Objective 1 Treatment</th>
<th>Time Point (min.)</th>
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<tbody>
<tr>
<td>1</td>
<td>1</td>
<td>15</td>
</tr>
<tr>
<td>2</td>
<td>1</td>
<td>60</td>
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<tr>
<td>3</td>
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**Results**
Positive replicates of PRRSV were found by PCR for all time points in Treatment 1, 2, and 3, with no significant differences observed for the number of positive replicates between treatments at any time point. No positive replicates were identified for Treatment 4. Polymerase chain reaction results for TGEV were more variable. Treatment 1 had fewer positive replicates than Treatment 3 at time points 2 and 4. A significant difference was not observed between Treatment 1 and Treatment 2 at any time point. All replicates in Treatment 4 tested negative for TGEV by PCR.
A significant decrease in PRRSV genomic copies by qPCR was noted between Treatment 1 and both Treatments 2 and 3 at time point 3. Additionally, Treatments 1 and 2 were significantly decreased in comparison to Treatment 3 at time point 4.

All 4 study pigs in Bioassay Group 6 tested positive for PRRSV by PCR at 7 and 14 days PI, and they were positive for PRRSV by ELISA at day 14 PI. No other bioassay groups tested positive for TGEV or PRRSV by PCR or ELISA at either sampling day. Transmissible gastroenteritis virus IHC was negative on all study pigs.

**Discussion**

The design of the washing procedure was successful in replicating features often found on commercially used livestock trailers, such as randomly scattered organic matter and water pooling. Results from this study show Accel disinfectant decreased the amount of PRRSV genomic copies on board the contaminated model trailers quicker than Synergize disinfectant or no disinfectant when paired with a 90-second cold-water, high-pressure wash. Both Accel and Synergize were effective at inactivating PRRSV by 15 minutes post-application of disinfectant. A cold-water, high-pressure wash alone was not effective at preventing inoculated bioassay pigs from becoming viremic with PRRSV with samples collected 15 minutes post-washing. The efficacy of Accel and Synergize against TGEV cannot be assessed in this study because of the lack of a PCR, ELISA, or IHC positive result for any pigs in Bioassay Group 5.

**References**
