Summary of: Evaluation of viral RNA extraction methods to detect PRRS and influenza A viruses from used commercial HVAC air filters from swine farms

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Key Points

- Little is known about the type and nature of viruses that get trapped in commercial filters on swine farms due to a lack of sampling methods.
- The first part of this study established effective methods for eluting and identifying PRRS and IAV viruses in MERV filters under lab conditions.
- The second part of the study tested the verified method on used filters from swine farms, verifying the presence of both PRRS and IAV

Porcine reproductive and respiratory syndrome (PRRS) and Influenza A virus (IAV) are both transmitted by air and are the two major respiratory viruses that often co-circulate in swine farms. Air filtration systems are used by swine farms as part of their biosecurity measures, implemented to reduce infection risk, often targeting PRRS. Over the last decade the number of filtered farms in the United States has increased drastically, particularly in swine dense regions. Despite the significant investments made in filtration systems, the nature and type of viruses trapped in these filters is not yet clear because of the lack of available and reliable methods to elute and detect these viruses. The main goal of this project was to evaluate the feasibility of identifying PRRS and IAV on used air filters retrieved from swine farms. First it was necessary to identify the best combination of sample elution and RNA extraction methods for detecting PRRS in MERV14 filters. The finalized process was then applied to used filters taken from midwestern farms in swine dense areas.

A pilot study was conducted in order to identify the best RNA extraction method. New MERV14 filters were spiked with the VR-2332 PRRSv reference strain in varying dilutions. Sections of the filters were then removed and used to test four eluting methods, elbow shake, vortex, freeze thaw, and liquid nitrogen grind. The vortex, freeze thaw, and liquid nitrogen grind methods were the most effective. These were then combined with two different RNA extraction strategies, namely MagMAX and TRIzol. The best combination of elution and RNA extraction was identified based on statistical significance for both, the cycle threshold (CT) RT-PCR values and RNA copies/ml. The liquid nitrogen grinding elution and TRIzol combination was the most effective.

This newly standardized extraction methodology was then used to test used HVAC commercial filters from swine barns. Filtration use is targeted primarily at stopping new PRRS infections, but the study opted to target both IAV and PRRS since both viruses are airborne and often co-circulate in farms. Samples were taken from 44 used MERV filters, originating from 13 different farms belonging to 4 production companies. All the farms were located in swine dense midwestern regions, had a history of PRRS infection prior to filtration, and the filters had been in use for 1-3 years.

The results of the 44 sampled filters showed that both PRRS and IAV were able to be identified in used filters from breeding farms. PRRS was identified in 27% (12/44) of the used filters from 31% (4/13) of the farms. IAV was identified in 66% (29/44) of the filters from 77% (10/13) of the farms. Samples that were positive for PRRS had CT values ranging from 32.21 to 34. 57, while IAV positive sample CT values ranged between 26.96 to 34.93. There was an unexpected finding that filters from two farms known to be PRRS negative at the time of filter removal, had filters that tested positive for PRRS. This suggests that the filters were able to prevent the introduction of viable PRRS viruses from entering and infecting the farm, while further supporting the possibility of airborne PRRS transmission.

This study continues to add evidence of aerosol spread together with providing a new approach to understanding airborne transmission as well as a potential monitoring methodology.

Table 6
Number of filters qRT-PCR positive and suspect for porcine reproductive and respiratory syndrome virus (PRRSv) and influenza A virus (IAV) by farm. The status of the farm for PRRS virus if known is shown and was provided by the producer or was obtained from the MSHMP database.

Farm code	No. filters/ farm	No. of PRRSv positive filters[a]	No. of filters suspected for PRRSv	No. filters positive for IAV	No. filters suspected for IAV	PRRSv farm status at time of filter removal
1	5	1	0	5	5	Positive
2	4	1	0	1	4	Positive
3	1	0	0	1	1	Positive
4	1	0	0	0	1	Positive
5	7	1	3	4	7	Negative
6	12	9	2	11	7	Negative
7	2	0	0	2	2	N/A
8	2	0	0	1	2	N/A
9	2	0	0	0	2	N/A
10	2	0	0	0	2	N/A
11	2	0	0	2	2	N/A
12	2	0	0	1	2	N/A
13	2	0	0	1	2	N/A
Total	44	12	5	29	39	

[a] Filters were considered positive if real-time RT-PCR cycle threshold (Ct) values were ≤35, suspect >35 and < 40, and negative if Ct ≥ 40.

Find the full article (possibly limited access) here: UMN PRRS and IAV filter testing article or by contacting Dr. Montse Torremorell at torr0033@umn.edu .