COMPARISON OF FILTER MATERIALS, WASH SOLUTIONS, AND EXTRACTION METHODS IN THE DETECTION AND QUANTIFICATION OF INFLUENZA VIRUS

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Background

Influenza is a contagious respiratory illness transmitted through the inhalation of aerosolized influenza virus.

Few researchers have detected airborne viruses in the field. New air sampling methods are needed to improve the detection and quantification of influenza virus aerosol.

A new industrial hygiene filter material made from polystyrene (PS) has been developed for the collection of bioaerosols. However, no data are available on the recovery of influenza virus from PS sampling media.

Aims

To compare influenza virus recovery from PS filter material to commonly used PTFE and PVC filter materials.

To compare the effect of air passing through the filter materials on virus recovery.

To compare virus removal from the filter materials using commonly used HBSS and PBS wash solutions.

To compare RNA extraction using an optimized Trizol method versus a commercially available extraction kit.

Methods

Filters were spiked with a known concentration of Influenza A H1N1, and placed into 37 mm closed face cassettes (n=240).

Experiments were conducted in a chamber for 30 minutes at a targeted 50% relative humidity (RH).

Six cassettes per experiment were used. Two of each type of filter were used [*i.e.*, Polytetrafluoroethylene (PTFE), polyvinylchloride (PVC), and polystyrene (PS)].

Three cassettes (one of each filter type) were treated with filtered chamber air at a rate of 4 liters per minute (LPM).

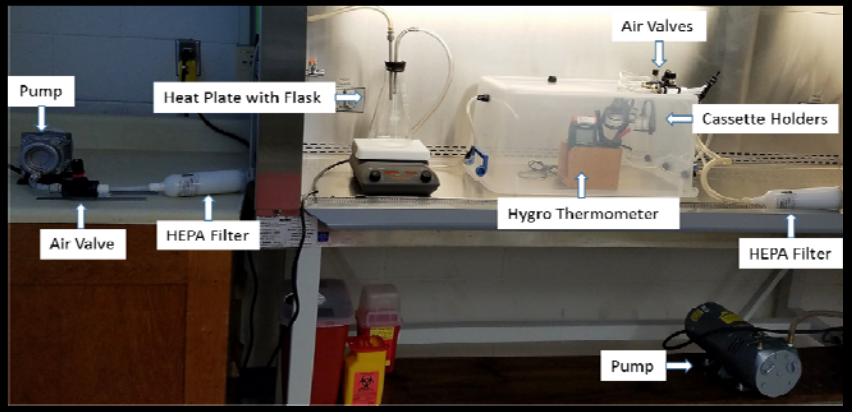
Filters were washed with phosphate buffered saline (PBS) or Hanks balanced salt solution (HBSS) to remove virus.

RNA was extracted using the Trizol method or QIAamp Viral RNA Mini Kit.

RNA was quantified and counted using RT-qPCR.

Data distribution was determined using Box-Cox Transformation, and data were analyzed using Mixed Effects Model in R.

Methods



Results

Geometric Mean (GSD) Influenza Virus Count

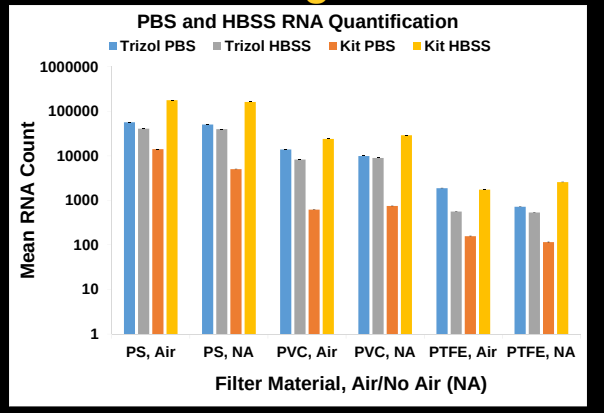
Extraction		Wash	Filter Type		
			PTFE*	PVC*	PS*
Air	Kit‡	PBS†	158 (3.81)	626 (7.00)	14,241 (7.11)
		HBSS†	1,755 (3.47)	24,155 (3.12)	179,443 (1.45)
	Trizol‡	PBS	1,898 (2.87)	13,954 (1.75)	57,176 (2.00)
		HBSS	564 (4.21)	8,325 (4.29)	41,164 (4.90)
No Air	Kit‡	PBS†	117 (5.14)	755 (8.27)	5,081 (2.83)
		HBSS†	2,599 (2.26)	29,220 (2.21)	162,897 (1.96)
	Trizol‡	PBS	726 (3.49)	9,939 (2.37)	51,020 (1.35)
		HBSS	537 (3.95)	9,057 (5.40)	39,942 (4.47)

*Significant difference in viral count between each filter material

† Significant difference in viral count between HBSS and PBS when using the kit extraction method

‡ Significant difference in viral count between kit and Trizol extraction methods, regardless of filter type

Background



Conclusions

The new PS filter material had the highest viral recovery, compared to PVC and PTFE.

Viral recovery varied based on extraction method, wash combination, and filter type.

HBSS wash combined with the QIAamp Viral RNA Mini Kit had higher virus recovery than the HBSS and Trizol combination.

PBS wash combined with the Trizol method had higher virus recovery than the PBS and QIAamp Viral RNA Mini Kit combination.

Future Research

Compare aerosolized virus recovery using the new PS filter material to other impinger or filter based bioaerosol samplers.

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