Sampling for Airborne Influenza Virus Using RNAPB: A New Approach

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Background

Characterizing airborne influenza virus exposure may be important for infection prevention and control.

Determining airborne concentrations of influenza virus is difficult due to degradation of viral RNA by RNAses and damage to virus structure inflicted during air sampling.

RNA preservation buffers (RNAPB) may protect against RNAses and stabilize even low levels of influenza virus for months at ambient conditions.

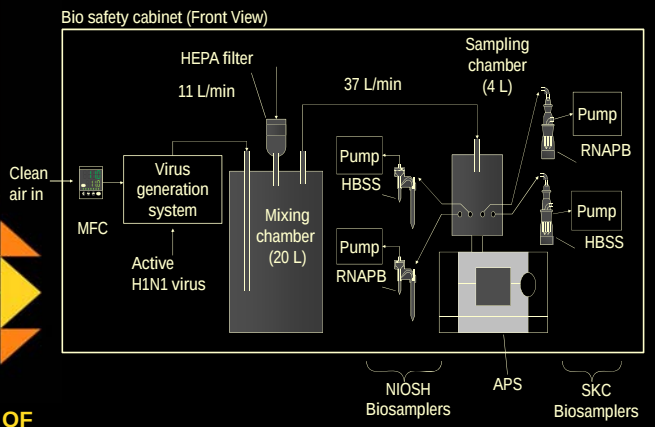
A biosampler was recently developed by NIOSH to capture airborne virus and was shown to do so as effectively as the "gold standard," SKC Biosampler.

Objectives

1) Compare ability of two bioaerosol samplers – SKC Biosampler and NIOSH Biosampler – to collect aerosolized virus
2) Compare effectiveness of RNAPB vs. HBSS as a sampling media for recovery of viral RNA
3) Evaluate ability of RNAPB to maintain viral RNA integrity at room temperature conditions vs. HBSS

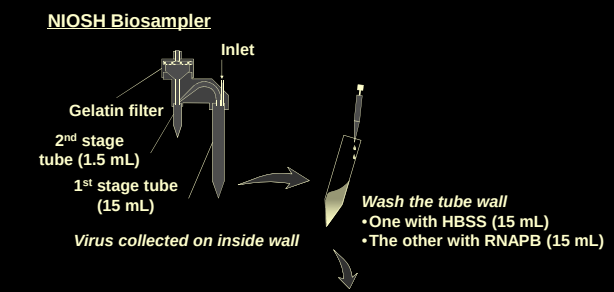
Experimental Setup

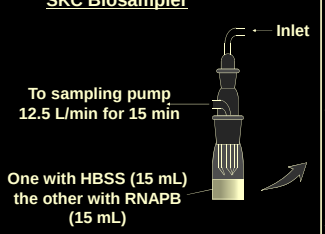
Figure 1: Aerosol generation, mixing, and sampling setup for 10 repeat experiments



Method

Figure 2: SKC and NIOSH Biosamplers and sample analysis





Real-time qPCR

1. Performed for NIOSH and SKC Biosamplers for each trial to measure initial concentration of viral RNA collected by samplers
2. Preservation of viral RNA in RNAPB vs. HBSS: three aliquots from each SKC sampler were stored at room temperature for 1, 4, 7, 14 days
3. Results were analyzed using a general linear model for a two-way ANOVA

Results

Objective 1:
The SKC Biosampler collected a higher concentration of virus particles / L air ($p<0.001$)

Objective 2:
Virus samples collected in HBSS measured a higher virus particle concentration than samples collected in RNAPB ($p<0.001$)

Table 1: Mean (SD) number of virus particles collected / L sampled air

SKC RNAPB	SKC HBSS	NIOSH RNAPB	NIOSH HBSS
30.4 (49.3)	215.4 (139.5)	0.3 (0.5)	12.3 (20.3)

Results, continued

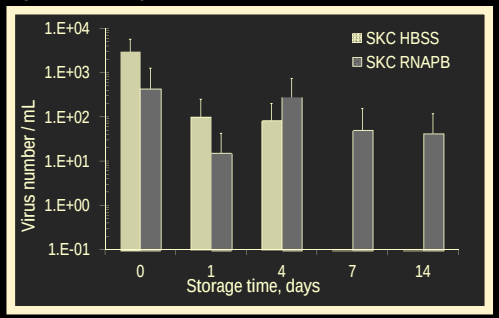
Objective 3:
The virus particles collected in HBSS had a mean starting virus concentration more than 7 X that of samples collected in RNAPB.

By day 4, the RNAPB samples had an average viral RNA concentration 3 X that of HBSS.

By day 7, RNAPB samples had an average of 500 X more viral RNA than the HBSS samples (49 particles/mL vs. < 1 particle/mL).

By day 14, RNAPB virus concentration had stabilized around 42 particles/mL, and no virus could be detected in HBSS samples.

Figure 3: Effect of media type on virus stability in room temperature storage, mean virus particles/mL of media



Conclusions

SKC Biosampler collected more virus than the NIOSH Biosampler.

HBSS measured more virus than RNAPB as a sampling matrix.

RNAPB preserved virus at concentrations up to 500 times higher than HBSS over a longer storage period.

RNAPB should be used as a sampling matrix if samples will be stored at room temperature for greater than 2 days (*e.g.*, long shipping times).

Improved extraction methods are needed to extract viral RNA from RNAPB.

Future Research

Sampling recommendations from this project are currently being applied to field work to quantify exposures of health care workers to airborne influenza.

Acknowledgements

Thanks to Drs. John Noti and Bill Lindsley at NIOSH for supplying us with the NIOSH biosampler. This research was supported by the National Institute for Environmental Health Sciences through the University of Iowa Environmental Health Sciences Research Center, NIEHS/NIH (P30 ES05605) and and by an ERC training grant from the Centers for Disease Control, National Institute of Occupational Safety and Health (T42OH008491).

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