Toward Identifying the Most Effective Samplers for Airborne Viruses

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Motivation

• Emerging zoonotic influenza viruses pose real or potential risks to swine, poultry, and veterinary workers
• Many viruses may be transmitted through air among animals or between animals and people, or have potential to develop transmissibility
• Animals in agricultural facilities generate virus-containing particles small enough to be transported substantial distances
• Little is known about typical concentrations and sizes of airborne virus-containing particles in animal agriculture, or if viruses remain infectious
Why do we care about particle size?

• We want to know how far virus-containing particles are able to travel through air
• We want to determine where virus-containing particles deposit in human or animal respiratory tract
• We want to identify technologies that can remove virus-containing particles from air
Research Objective

Identify/develop a high-volume, field-portable, size-differentiating viral aerosol sampler and use it to measure worker exposures to live airborne influenza viruses in animal agriculture facilities.

- We want large samples to achieve low limits of detection.
- We want to do this in the real world.
- Our focus is animal agriculture.
- We're working with viruses.
- The particles that we're considering are airborne.
- We're collecting samples from the air.
- We want to know if the viruses in the air are infectious.

Hey...we already talked about this.
First Step: Evaluate Existing Samplers

• Assemble wide range of existing samplers that collect viral aerosols by variety of principles
• Test samplers side-by-side in an isolation room using mechanically-generated influenza virus aerosols
• Determine combinations of sampling parameters and technologies that collect greatest quantity of viral RNA and live virus
Sampling Technologies

- Impingers
- Cyclones
- Impactors
- Filters
- Electrostatic collection
- Combinations

### Samplers Evaluated

<table>
<thead>
<tr>
<th>Group 1</th>
<th>Group 2</th>
<th>Group 3</th>
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<tbody>
<tr>
<td>Non-Viable Andersen Cascade Impactor (ThermoFisher)*</td>
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<tr>
<td>Cyclonic Collector (Midwest Micro-Tek)*</td>
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<td>Cyclonic Collector (Midwest Micro-Tek)*</td>
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<tr>
<td>AGI-30 impinger (Ace Glass, Inc.)</td>
<td>47mm fiberglass filter</td>
<td>MOUDI (MSP Corp.)</td>
</tr>
<tr>
<td>BioSampler (SKC Inc.)</td>
<td>47mm gelatin filter</td>
<td>Trichotomous Virtual Impactor Sampler</td>
</tr>
<tr>
<td>Cyclone Bioaerosol Sampler (NIOSH)</td>
<td>PEMS PM2.5 sampler (SKC Inc.)</td>
<td>University of Minnesota)</td>
</tr>
<tr>
<td>SpinCon II (InnovaPrep)</td>
<td>Hi-Vol TSP sampler</td>
<td>Series 230 High Volume Cascade Impactor</td>
</tr>
<tr>
<td>Bobcat (InnovaPrep)</td>
<td>Electrostatic sampler</td>
<td>(Tisch Environmental)</td>
</tr>
<tr>
<td>VIVAS (UF &amp; Aerosol Dynamics)</td>
<td>(UNC-Chapel Hill)</td>
<td></td>
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</tbody>
</table>

*Sampler was used in all three groups as a control*
Methods

• H3N2 swine influenza virus (SIV) grown and titrated in Madin-Darby canine kidney (MDCK) cells grown in Eagle’s MEM with supplements
• Fluorescein dye added to virus suspensions to track physical collection efficiency
• SIV suspension aerosolized at pressure of 20 psi using 6-jet Collison-type nebulizer in an isolation room in the BSL-2 Veterinary Isolation Building on University of Minnesota St. Paul campus
• Simultaneous samples collected by samplers in each group for 30 minutes
• Samplers were tested in three replicate tests
• Resulting nebulizer suspensions and air samples analyzed
  – SIV titrated to determine quantities of live virus
  – Viral RNA extracted and used for qRT-PCR (quantitative real time-PCR) to determine quantities of total virus
  – Intensity of fluorescein dye measured by spectrofluorometry
• Relative recovery calculated to determine fraction of collected virus still active
• Recoveries among the samplers were compared descriptively and statistically
Isolation Room Setup
Live Virus Titer, Set #1

![Bar chart showing geometric mean live virus titer (TCID50/mL) for different samplers: Andersen Impactor, Cyclonic Collector, AGI-30, BioSampler, NIOSH, Spin Con II, Bobcat, VIVAS. Each bar represents the mean titer with error bars indicating variability.](umash.umn.edu)
Live Virus Sampled, Set #1

![Bar chart showing the geometric mean live virus sampled (TCID50) for different samplers.](image_url)
Live Virus Air Concentration, Set #1

![Graph showing geometric mean live virus air concentration (TCID50/m³) for different samplers](umash.umn.edu)
Total Virus Observed, Set #1

Geometric Mean Total Virus Observed (RNA copies/mL)

- Andersen Impactor
- Cyclonic Collector
- AGI-30
- BioSampler
- NIOSH
- Spin Con II
- Bobcat
- VIVAS
Total Virus Air Concentration, Set #1

Geo Mean Total Virus Air Concentration (RNA copies/m$^3$)

- Andersen Impactor
- Cyclonic Collector
- AGI-30
- BioSampler
- NIOSH
- Spin Con II
- Bobcat
- VIVAS

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Relative Recovery, Set #1

Andersen Impactor
Cyclonic Collector
AGI-30
BioSampler
NIOSH
Spin Con II
Bobcat
VIVAS

Geometric Mean Fluorescein Relative Recovery
Total Virus Observed, All Sets

![Graph showing geometric mean total virus observed for different samplers. The x-axis represents samplers, and the y-axis represents RNA copies/mL. Different samplers have bars indicating the mean and error bars showing variability.]
Total Virus Air Concentration, All Sets

Geo Mean Total Virus Air Concentration (RNA copies/m³)

Samplers

Andersen Impactor, Cyclonic Collector, AGF-30 Biobase, NIOSH Spin Con, Bobcat, VIVAS Andersen Impactor, Cyclonic Collector, Glass Fiber Filter, Gelatin Filter, PEMS Hi-Vol TSP Filter, Electrostatic-Sampler, Andersen Impactor, Cyclonic Collector, MOUDI Impactor, Trichotomous Sampler, Series 250 Impactor
Discussion

• High flow rate samplers tend to yield higher titers/more RNA copies
  – High flow samplers consolidate sample more than lower flow samplers
  – Likely better for detection of airborne viruses at low concentrations

• Highest airborne virus concentrations observed among lower flow rate samplers
Discussion (continued)

• Impinger samplers may keep virus live more effectively than other types of samplers
• Ease of use important but should not drive decisions
• Two-sampler strategy may have benefits during outbreak investigations
  – High flow, non-sizing sampler for detection
  – Lower flow, size-separating sampler for concentration measurements
Bottom Line
No sampler that we have tested is “best” so far

Next Steps
• Compare several of best-performing samplers in field tests this flu season
• Design and build novel size-separating sampler
• Compare novel sampler to existing ones
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