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Comparison of high-volume air sampling equipment for viral aerosol sampling during emergency response

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ABSTRACT

Objective: This study compared the performance of two high-volume bioaerosol air samplers for viable virus to an accepted standard low-volume sampler. In typical bioaerosol emergency response scenarios, high-volume sampling is essential for the low infective concentrations and large air volumes involved.

Design: Two high-volume air samplers (XMX/2L-MIL and DFU-1000) were evaluated alongside a low-volume sample (BioSampler). Low and high concentrations (9.3-93.2 agent containing particles per liter of air [ACPLA]) of male-specific coliphage 2 (MS2) virus were released into a 12 m^3 aerosol test chamber and collected using the air samplers. The collection media from the samplers were then processed and viable virus was assessed via plaque assay.

Setting: Aerosol test chamber.

Subjects, participants: None.

Interventions: Collection media and flow rate were modified for the XMX/2L-MIL sampler for viable analysis.

Main outcome measures: Concentration estimates in units of plaque forming units per liter of air (PFU/liter) assessed by the samplers as compared to the levels inside the chamber as evaluated with a slit to agar plate in units of ACPLA. Comparison was made via one-way analysis of variance.

Results: Both the XMX/2L-MIL and DFU-1000 achieved collection effectiveness equal to or greater than the low-volume air sampler for the evaluated MS2 concentrations. The XMX/2L-MIL reliably collected quantifiable low concentrations of MS2, but the DFU-1000 was unable to do so.

Conclusions: For emergency response to suspected bioaerosols, the evaluated high-volume samplers are as effective as the standard low-flow sampler and should be considered in conducting a health risk assessment. If low concentrations are expected, then high-flow samplers using liquid collection are preferred.

Key words: bioaerosol sampling, bacteriophage, virtual impactor, bioterrorism

INTRODUCTION

Outbreaks of airborne viral disease represent one of the greatest emerging risks to public health. High priority viral agents include influenza, variola, viral hemorrhagic fevers, and equine encephalitis.[1,2] Resources from government and private agencies have been allocated to address these risks; however, actions taken thus far have failed to meet national requirements to adequately prevent casualties during an incident involving a biological agent. The Commission on the Prevention of Weapons of Mass Destruction Proliferation and Terrorism chartered by the United

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States Congress issued a "grade of F" on current efforts to "enhance the nation's capabilities for rapid response to prevent biological attacks for inflicting mass casualties."[3]

Health risk assessment of an aerosolized viral agent requires the accurate detection, identification, and quantification of viral aerosols. This need is complicated by the urgency with which information is often required and the dilute concentrations expected during an environmental release of a viral agent. To meet the needs for fast response and to provide a sufficient quantity for analysis, high-volume air sampling equipment is typically used in an emergency response to a biological agent. Detection and quantification of an airborne virus is dependent on the concentration of virus present, collection efficiencies of the air sampling methodology, and the sensitivity of the sample analysis method used.[4] However, few high-volume air samplers have been rigorously evaluated. Numerous commercially available low-volume air samplers have been evaluated and are effective at collecting viral aerosols and can be used as a benchmark for high-volume samplers. Examples include solid impaction, liquid impinger, swirling aerosol collection, and slit to agar (STA) impaction.[5]

Two high-volume bioaerosol samplers available in civil and defense inventories are the XMX/2L-MIL and DFU-1000. The XMX/2L-MIL and DFU-1000 have been used in field studies to sample for viral aerosols. The XMX/2L-MIL was used in 2004 to sample areas contaminated with H7N3 avian influenza and was capable of collecting detectable quantities of virus in a sample when analyzed with real-time polymerase chain reaction (RT-PCR).[6] The DFU-1000 was used in a 2006 study attempting to sample areas where adenovirus-4 was suspected and was capable of collecting samples with detectable levels of adenovirus-4 when the samples were analyzed with PCR.[7] However, PCR does not give a direct measure of quantification such as viability does. The capability of the XMX/2L-MIL and DFU-1000 to recover viable virus for analysis cannot be assessed from these studies.

The use of air sampling equipment in an emergency response setting must be justified and balanced with other requirements throughout a response. The use of high-volume air sampling equipment in a response environment would typically require dedicated and specialized entry personnel. Therefore, high-volume air sampling equipment must be capable of detecting and identifying aerosolized biological agent and providing actionable health risk information to incident commanders and other decision-making authorities. The use of high-volume air sampling equipment in conjunction with PCR is capable of producing agent detection and identification results within 1 hour following sample collection. In situations where viable analysis is required to assess the health risk of a released agent, sample results could require up to 24 hours for analysis. Viable samples can be used as confirmation of PCR results.

Collection media

Previous low-flow impinger studies conducted to optimize liquid media for the collection of Porcine Reproductive Respiratory Syndrome Virus have shown preservative media has similar capabilities to phosphate buffer saline (PBS) solution in the collection of viral aerosols. Preservative liquid media is preferred to PBS in situations where long-distance transport or significant sample hold times may be necessary. However, preservative media can "foam" at high sample flow rates in liquid impingers resulting in a significant loss of media. The XMX/2L-MIL manufacturer validated its effectiveness for aerosolized male-specific coliphage 2 (MS2) in an unpublished study using PBS, but not preservative media. Preservative media was desired for this study because of the ability to keep viruses viable when sample hold times extend beyond a few hours. These situations are common in deployed locations between response collection points and definitive biological laboratories.

Aerosol test chambers

Using an aerosol test chamber (ATC) permits initial evaluation of a sampling method in a controlled environment with known virus concentrations in air.[5] Many studies also use nonpathogenic surrogate viruses to simulate pathogenic viruses with similar aerodynamic and chemodynamic characteristics to preclude expensive biosafety precautions.[8] Bacteriophages are

often used as viral surrogates in aerosol studies, including MS2.

METHODOLOGY

High-volume air sampling equipment

Two high-volume air sampling systems were selected for evaluation from existing United States Department of Defense inventories. These systems included a dry filter collection system, the DFU-1000 (Lockheed Martin Integrated Technologies, Springfield, VA), and a high-volume air sampling system incorporating a multistage virtual impaction module combined with a liquid impinger, the XMX/2L-MIL (Dycor Technologies, Edmonton, Alberta, Canada). Both systems were specifically designed for the collection of aerosolized biological warfare agents and are currently deployed in a variety of applications in both biological defense and biological monitoring. The DFU-1000 weighs 19 kg and is 38 cm high, 33 cm wide, and 33 cm deep. It uses a 5.6 A, 120 V common power source.[9] The XMX/2L-MILweighs 17 kg and is 58 cm high, 46 cm wide, and 33 cm deep. It uses a 10 A, 110 V power source.[10] These systems were designed to operate at very high air flow rates. Two copies of the same XMX/2L-MIL model were operated at 620 liters per minute (Lpm) to collect air samples using a virtual impactor with final collection through a liquid impinger. The DFU-1000 was operated at 760 Lpm to collect air samples using two 1.0-μm-sized polyester felt filters. Flow rates for the high-volume air sampling equipment were verified with an anemometer. Air samplers were allowed to sample from the ATC for 5 minutes per trial. The XMX/2L-MIL uses a multistage virtual impactor to concentrate collected particles from a high-volume flow stream into a low-volume flow stream of approximately 13.5 Lpm. This low flow stream is then transferred into a liquid impinger for final collection. For this study, the flow rate into the liquid impinger was reduced by insertion of a flow reducer to approximately 4 Lpm to prevent foaming and loss of the preservative media volume and biasing the estimate of concentration.

Low-volume sampling equipment

The BioSampler (SKC, Eighty Four, PA) was used as a low-volume benchmark in the comparison of the high-volume air samplers. This sampler was chosen due to its widespread use in previously published studies and proven effectiveness in the collection of micron- and submicron-sized airborne particles. The BioSampler has greater than 65 percent collection efficiency for 0.3-μm-sized particles and greater than 70 percent collection efficiency for 1.0-μm-sized particles.[11] The BioSampler was attached to a stand inside the chamber within 40 cm of the DFU-1000 sample port and raised to the inlet height of the DFU and XMXs. The BioSampler was operated at 12.5 Lpm using an SKC Vac-U-Go noncompensating vacuum pump (SKC) and was calibrated before and after sampling each day using a Bios DryCal DC-2 Air Flow Calibrator (Bios International, Butler, NJ). A field rotameter (SKC) was used with the BioSampler as a secondary flow standard to verify maintenance of airflow during the trials.

Collection media

Remel MicroTest M5 Multi-Microbe Media, "M5 media" (Remel, Lenexa, KS) was used as the collection media for all instruments in this study. M5 media is typically used in the transport of clinical samples containing viruses and other pathogens and maintains agent infectivity. M5 media had not been used in the collection of environmental air samples but should offer similar advantages in maintaining the viability of a viral agent. M5 media consists of a mixture of Hank's salt solution, bovine serum albumin, protein stabilizers, sucrose, glutamic acid, phenol red, antimicrobial, and antifungal additives. Following manufacturer specifications, 5 mL of liquid media was added to the XMX/2L-MIL collection vessel prior to sampling and 20 mL was added to the BioSampler. To ensure uniformity between sampling methodologies, M5 media was also used in the liquid extraction of sample from the polyester felt filter collected from the DFU-1000 described later. The tendency of protein and carbohydrate media to foam required a 0.1 percent concentration of antifoam (Y-30 Aqueous Emulsion, Sigma Aldrich, St. Louis, MO) be added to the liquid media prior to sampling.

Aerosol test chamber

Aerosol dispersion and collection were conducted in a flow-through ATC provided by Dycor Technologies

Ltd. The ATC was 12 m^3 in volume (3 m × 2 m × 2 m). Four cubic meters of High Efficiency Particulate Air (HEPA)-filtered air per minute was supplied to the chamber. This airflow combined with the 0.2 m^3/min of airflow from the aerosol dispersal pipe creating a nominal air velocity through the chamber of approximately 1.75 cm/s.

Three high-volume sample ports were located approximately 1.9 m from the point of aerosol dispersal. The chamber was equipped with two circulating fans for aerosol mixing. The inlets of the two XMX/2L-MIL samplers were raised into the ATC to a height of approximately 16 cm. The DFU-1000 was attached to a 5-cm-diameter copper pipe raised into the chamber to a height of 16 cm, consistent with the two XMX/2L-MIL samplers. Intake height was kept constant among these instruments to eliminate bias in sampling that might occur by differences in sample collection height. A diagram showing the inside of the ATC is given in Figure 1. Temperature was maintained within a range of 25.7-26.3°C, with a relative humidity range of 30.5-34.3 percent. Following each sample period, the ATC was purged thoroughly with clean air.

Surrogate viral agent MS2

MS2 (American Type Culture Collection [ATCC] 15597-B1, Manassas, VA) was the surrogate viral agent for this study. MS2 offers several advantages not offered by other agent species. Male-specific coliphages such as MS2 can be controlled in the laboratory and are readily aerosolized. MS2 is an icosahedral unenveloped RNA bacteriophage of the *Escherichia coli* bacteria and is 27-34 nm in size[12] and has been used as a surrogate agent for previous studies.[8,13] Additionally MS2 has a reasonably high stability when aerosolized, with previous studies showing up to 52 percent of aerosolized MS2 remaining viable 45 minutes after aerosolization.[14]

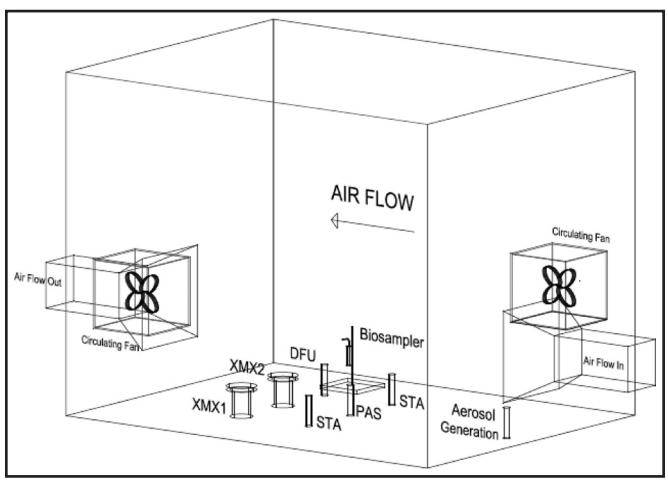

Figure 1. Interior view of aerosol test chamber depicting air intakes and outlets, aerosol generation port, circulating fan locations, locations of high- and low-volume air sampling points, and location of slit to agar (STA) and location of portable aerosol spectrometer (PAS).

Aerosolization of MS2

MS2 solution was aerosolized using a Sono-Tek 8700-48MS ultrasonic atomizing nozzle (Sono-Tek, Milton, NY) mixed with 200 Lpm of HEPA-filtered air and introduced into the chamber. Final aerosol concentration was determined using STA plaque counting. Two STA biological air samplers, model number STA-203 (New Brunswick Scientific, Edison, NJ) were used for this purpose. These STA samplers were operated at 30 Lpm to impact particle sizes greater than 0.5 μm onto a rotating plate.[15] The STA sampler gave a measure of viral particle load in agent containing particles per liter of air (ACPLA). As the slit rotated around the agar plate, particles from the chamber impacted. This is not perfectly correlated to the air sampler instrument performance. A particle may have many viruses within it, and higher concentrations in the chamber tend to have larger particles with more viruses per particle. The STA sampler will still only indicate a single plaque for that particle at its impact point. By contrast, the low- and high-volume air samplers being evaluated were used so that the agent containing particles were transferred to a preservative liquid media and the media deposited across an agar plate. This may allow a single large particle to contribute many viruses for plaques in the analysis. The resulting concentration unit for the air samplers was plaque forming units per liter of air (PFU/liter).

Particle size measurement

Particle size characteristics were recorded continuously for each sample run using a Portable Aerosol

Spectrometer (PAS) Model 1.108 (Grimm Technologies, Douglasville, GA). This PAS provided size and distribution data for particles ranging from 0.3 to 20 μm in size. This monitoring allowed for additional MS2 aerosol to be injected to the chamber as required for maintaining MS2 concentration.

Extraction of MS2 sample from DFU-1000 filter

Each of the 46 mm filters was removed from the DFU-1000 filter inserts and placed in a 50 mL collection tube with 15 mL of M5 media. The media was allowed to make complete contact with the filter material by hand agitating the collection tube immediately after collection. Once received in the laboratory, the filters and sample media were vortexed for 10 seconds at 3,200 rpm allowing the collected sample to separate from the filter into the media for analysis.

Viable culture analysis

Air samples were analyzed for infectious virus using plaque assays.[16] Plaque assay plates were prepared using 10 mL of MS2 growth media with an overlay of 200 μL of *E. coli*, ATCC 15597 (8.75×10^8 colony forming units per milliliter [CFU/mL]) and 200 μL of sample. The overlay was mixed by lightly swirling the prepared plate. STA plates were prepared using 25 mL of MS2 growth media, with 500 μL of *E. coli* as an overlay. The MS2 growth media consisted of 5.6 g bacto-agar, 6 g of sodium chloride, 5 g of proteose peptone, 2 g of yeast extract, and 5.2 mL of glycerol. Plates were incubated overnight, for a minimum of 12 hours, at a temperature of 37°C.

Comparison of high-volume air sampling operating configuration

Previous unpublished studies conducted by Dycor Technologies collected MS2 bacteriophage using an unmodified XMX (13.5 Lpm secondary flow rate) with phosphate buffered saline collection media in the same ATC. As previously described, the XMX/2L-MIL systems used in the current study were modified by reducing the airflow to the impinger and using M5 media instead of phosphate buffer solution. In the previously conducted study by Dycor, five samples were collected using a single XMX over a relatively narrow viral chamber load range of 23.0-28.1 ACPLA. The results from this previous study in PFU/liter were standardized to units of PFU/liter per ACPLA present in the chamber. This allows for comparison with the results from the current study.

RESULTS

Collection of infectious MS2 bacteriophage

Overall results for the collection of aerosolized MS2 bacteriophage as a function of viral load present in the chamber are shown in Figure 2. Viral load in the chamber is expressed in terms of ACPLA.

As Figure 2 shows, the XMX/2L-MIL was as effective as the BioSampler low-volume air sampler to which it was compared. A large degree of intrainstrument variability was observed in the two XMX/2L-MIL samplers at high MS2 concentrations. The DFU-1000 demonstrated similar MS2 collection performance to the other evaluated air samplers at higher concentrations of MS2. At lower concentrations, the DFU-1000 underperformed the XMX/2L-MIL but was statistically similar to the BioSampler. Air sample results were then standardized by the viral load (ACPLA) present in the ATC to PFU/liter per ACPLA. This standardization allowed for a median MS2 air sample concentration to be determined using data collected from multiple levels of airborne viral load present in the chamber. Standardized results and median values are shown in Figure 3.

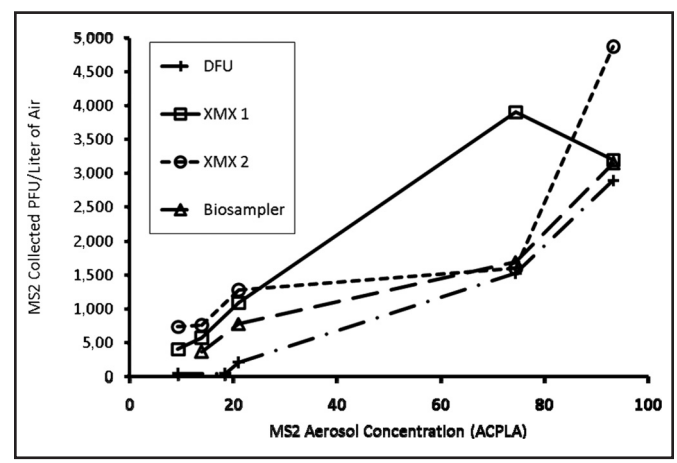

Figure 2. MS2 air sample collection relative to ACPLA present in chamber as measured by slit to agar sampler.

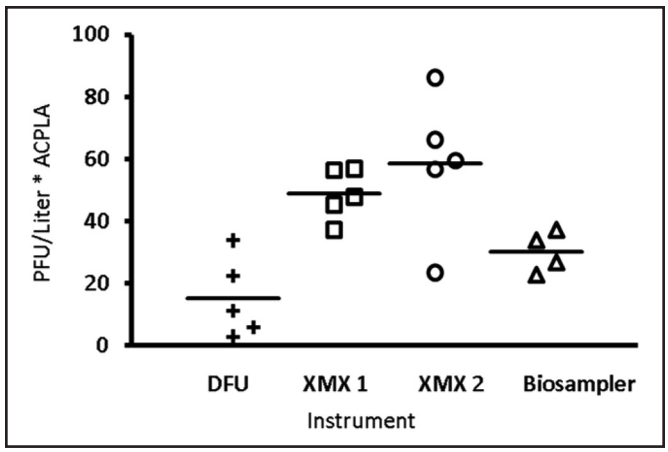

Figure 3. Air sample results per one ACPLA of MS2 aerosol in chamber.

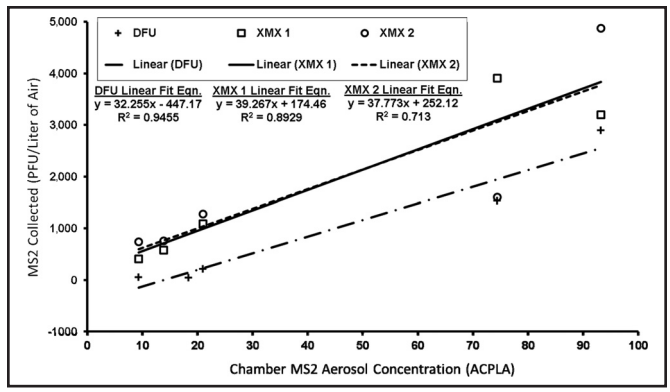

Figure 4. MS2 collection by high-volume air sampling equipment with linear trend lines.

Comparative analysis between all evaluated samplers using Kruskal-Wallis one-way analysis of variance (ANOVA; $\alpha = 0.05$) revealed a significant difference ($p = 0.009$) in the median standardized MS2 collection. A Dunn's rank sum post-test revealed the quantity of MS2 obtained using the DFU-1000 was significantly lower than quantity of MS2 obtained from the XMX/2L-MIL. This significant difference was found for both XMX/2L-MIL devices. Other comparison results using the Dunn's post-test reveal no significant difference between the XMX/2L-MIL and the BioSampler, or between the DFU-1000 and the BioSampler.

Quantification limitations of air sampling instruments. The lowest level of viral load produced in the chamber was 9.3 ACPLA. MS2 sample collection relative to ACPLA present in the chamber for the high-volume air samplers with linear trend lines is in Figure 4.

As indicated by the linear trend line, the DFU-1000 was unable to reliably deliver quantifiable results at MS2 concentration levels lower than approximately 14 ACPLA, although the higher degree of linear fit ($R^2 = 0.95$) indicates the DFU-1000 is capable of quantifying viruses present at higher concentrations. MS2 collection using the XMX/2L-MIL suggests a possible linear relationship relative to ACPLA present in the chamber. This allowed the XMX/2L-MIL to reliably produce quantifiable results for MS2 collection at the levels of airborne viral load evaluated in this project. A high degree of variability was observed between XMX/2L-MIL 1 and XMX/2L-MIL 2 at high MS2 concentrations ($R^2 = 0.89$ and 0.71, respectively).

Validation of preservative sampling media

The current study using XMX/2L-MIL air samplers and Remel media was compared to the previous unpublished Dycor study using XMX air samplers and phosphate buffer solution. The XMX model from the unpublished study used the same sampling components, flow rates, and procedures as the XMX/2L-MIL models in the current study. Standardized results (PFU/liter ACPLA) are shown in Table 1.

Data analysis using a Kruskal-Wallis one-way ANOVA ($\alpha = 0.05$) revealed a significant difference ($p = 0.027$) in the MS2 collection effectiveness between the two XMX operating methodologies. These results indicate a significant difference between the two configurations of the XMX. The reduction of impinger flow to approximately 4.0 Lpm and the use of preservative media combined were significantly more effective in the collection of viable MS2 than the standard operating configuration using full impinger flow of approximately 13.5 Lpm and PBS media. Either the use of airflow reduction to the impinger or the use of preservative M5 media, or both, could have been responsible for the differences in MS2 collection observed between the two studies.

MS2 aerosol particle size characteristics

Particle size characteristics are shown in Figure 5 grouped by MS2 concentration for each trial. Particle

Table 1. Comparison of XMX using PBS media and 13.5 Lpm of impinger flow (manufacturer specifications) with XMX using M5 media and impinger flow reduction to 4 Lpm (current study specifications) for MS2 concentrations

Instrument	No. of trials	Median sampled MS2 concentration per ACPLA present in chamber (PFU/liter ACPLA)	Range of sampled MS2 concentration per ACPLA present in chamber (PFU/liter ACPLA)
XMX operated using manufacturer specifications	5	36.30	32.90-38.78
XMX operated using current study specifications	10	56.55	37.21-79.57

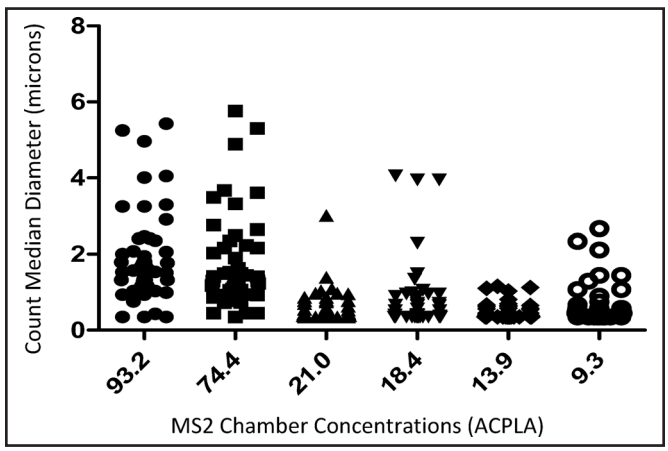

Figure 5. Count median diameter measurements grouped by trial MS2 concentration.

size characteristics were monitored every 6 seconds throughout the 5-minute trials and are presented in Figure 5 by the count median diameters (CMD) for each 6-second measurement grouped by trial MS2 concentration.

The high MS2 concentration trials, 93.2 ACPLA and 74.4 ACPLA, contained much larger concentrations of larger particles than the low MS2 concentration trials. The median CMD for the 93.2 and 74.4 ACPLA trials were 1.41 and 1.23 μm, respectively. The median CMD for the 21.0, 18.4, 13.9, and 9.3 ACPLA trials were 0.59, 0.62, 0.49, and 0.64 μm, respectively. These measurements indicate a significant difference in median particle size between trials where high and low concentrations of MS2 were used. As Figure 5 shows, significant variation was observed in the aerosol CMD measured by the PAS. This variation was not observed to be time dependent throughout any of the 5 minute sample periods and was likely caused by the injection of additional MS2 into the chamber as needed by the PAS feedback loop described earlier.

DISCUSSION

This study evaluated two high-volume air samplers in response to a surrogate viral agent, MS2 bacteriophage. Criteria evaluated included overall effectiveness of virus recovery over a range of airborne viral agent concentrations and an evaluation of virus preserving media in a high-volume air sampling system. Limitations of this research include the inability to achieve low agent concentrations representative of minimally infective doses, the relatively small number of samples collected and absence of repeat samples, and the inherent limitations in the use of a surrogate viral agent. The inhaled infectious dose of some viral agents such as Variola is estimated to be as low as 4 PFU based on studies using *vaccinia* virus in rabbits.[17] Further, the fluid dynamic interaction of the several samplers all inside the single 12 m^3 ATC was not characterized. It is possible that the high-volume samplers overwhelmed the low-volume sampler. However, the ATC did have two fans to keep the aerosol well mixed, and a PAS feedback for aerosol concentration.

This study demonstrated that the XMX/2L-MIL was capable of significantly ($p = 0.009$) greater collection of viable MS2 than the DFU-1000 under the sampling conditions evaluated for low MS2 concentrations (≤21.0 ACPLA). The collection of MS2 bacteriophage per volume of air sampled by the XMX/2L-MIL and

the DFU-1000 was also statistically similar to the collection of MS2 by the BioSampler. This study confirmed earlier observations in studies where collection of viral agent was attempted using both dry filter and liquid collection. Previous studies, such as a smallpox study conducted during the 1960s in India demonstrated methods using dry media recovered less viable virus than liquid collection methods.[18] Prior studies have also demonstrated significantly higher relative recoveries of aerosolized MS2 using liquid collection methods compared to the relative recovery obtained using dry nuclepore filters.[19] This study confirmed these earlier observations by showing the overall collection was significantly less for high-volume sampling methods using dry media than high-volume methods using liquid collection at lower chamber concentrations.

As also demonstrated by previous studies to optimize aerosol collection systems for viruses, collection media for liquid impinger sampling systems can have a significant influence on overall collection performance.[4] The results from this study showed the M5 media used in an XMX/2L-MIL with impinger flow reduction was very capable at collecting aerosolized MS2. While a statistically significant difference in MS2 collection between the modified XMX/2L-MIL apparatus used in the study and the standard XMX configuration was shown, there are several limitations to this comparison. First, the significance of the individual modifications within the systems cannot be assessed. Either the reduction in flow rate to the liquid impinger or the use of M5 media instead of PBS solution could have been responsible for the increased MS2 collection. A 2012 study showed significantly higher MS2 viable analytical results from M5 media as compared to PBS when operated at the reduced flow rate.[20] This suggests that the media is an important factor. However, as the M5 media cannot be used at the standard 12.5 Lpm flow rate, the flow rates cannot be compared across media. Second, the previous work conducted by Dycor evaluated the collection of MS2 by the XMX over a very narrow MS2 concentration range. The results from the current study were obtained over a wider range of MS2 concentration in the chamber. Although results were standardized to 1 ACPLA to allow comparison over different concentrations, some difference may be attributable to variations in MS2 concentration between the two studies. The results of this current study nonetheless demonstrated M5 media with impinger flow reduction can be effectively used in viral air sampling applications with the XMX/2L-MIL.

The disparity in particle size between high and low MS2 concentration trials was indicative of the agglomeration occurring among particles containing viral agents. The XMX/2L-MIL uses virtual impaction to reduce the collection of submicron-sized particles from the flow stream before final capture with a liquid impinger. The relatively similar performance of the XMX/2L-MIL in the collection of MS2 to the low-volume BioSampler suggests submicron-sized particles were not overwhelming contributors of viable MS2 in the agent containing test aerosol. The virtual impaction module of the XMX/2L-MIL eliminates most submicron particles from the impinger flow stream; therefore, the BioSampler would be expected to significantly outperform the XMX/2L-MIL if significant amounts of viable MS2 were present in the submicron particles at the point of collection. The removal of submicron particles by air sampling systems using virtual impaction has been documented in previous studies.[21] A study has been conducted to determine the aerosol size-selective sampling characteristics of the XMX/2L-MIL with and without the low-flow adapter.[22] That study evaluated the XMX/2L-MIL capture and retention efficiency at 4.5 Lpm secondary flow within two different impinger fluids (PBS and Remel M5) using four sizes of fluorescent spheres: 0.7, 1.0, 1.9, and 3.1 μm. There was no significant difference between PBS and M5 for capture and retention efficiency at 4.5 Lpm. The XMX/2L-MIL had a capture and retention efficiency of approximately 20 percent for 0.7 μm particles as compared to the BioSamplers efficiency of 65-70 percent anticipated in that size range.[11] The XMX/2L-MIL was able to capture submicron particles with the modified conditions, but at lower efficiency than the BioSampler.

While this study demonstrated the XMX/2L-MIL was effective in the collection of MS2 when compared to a low-volume air sampler benchmark, the DFU-1000 was shown to be less effective than the XMX/2L-MIL.

This performance difference was particularly large for samples with lower MS2 concentrations (≤21.0 ACPLA). The two most likely causes of the relatively low MS2 collection by the DFU-1000 at low concentration levels are problems with filter extraction and desiccation, or drying of the MS2 during collection. Desiccation of the collected MS2 would reduce MS2 viability and aggregation of MS2 into larger particles during the high concentration trials could have allowed MS2 to withstand the desiccation process of airflow across the filter. Filter extraction is also a likely contributor to significantly reduced DFU-1000 performance at lower MS2 concentration levels. As the surface of the filter became sufficiently coated with MS2 containing particles, transfer of MS2 into liquid sample media during extraction would be expected to increase. Relatively low air concentration levels would be expected during a response to a viral agent and this study demonstrated the XMX/2L-MIL would likely outperform the DFU-1000 under such conditions. Linear regression of the plaque assay results for the MS2 collected by the DFU-1000 suggests MS2 concentrations could not be reliably quantified at levels below 14 ACPLA, which is significantly above the airborne concentrations required to infect a population for most viral agents of concern. However, the XMX/2L-MIL and the DFU-1000 were both effective in recovering detectable quantities of viable MS2 bacteriophage at all evaluated aerosol concentrations of MS2.

CONCLUSIONS

This study demonstrated that commercially available high-volume air sampling equipment can be effectively used to detect aerosolized viral agents. This study also suggested that high-volume air sampling equipment using liquid collection methods was capable of quantifying aerosolized viral agents at levels comparable to more established low-volume liquid collection methods. While the XMX/2L-MIL was shown to be more effective than the DFU-1000 in the collection of low concentrations of aerosolized MS2, this study did not provide an evaluation of the effectiveness of the XMX/2L-MIL at very low levels of airborne agent concentration. It cannot be inferred from this study that a nondetection result obtained from an air sample collected using the XMX/2L-MIL would indicate an acceptable level of airborne virus infectivity risk following an aerosol release of a viral agent. A comparison of the DFU-1000, XMX/2L-MIL, and other high-volume bioaerosol sampling systems at minimally achievable aerosol concentrations should be completed in future studies. This would allow for the use of high-volume air sampling methodologies to reliably determine exposure risk to a vulnerable population following the release of a viral agent. Air sample results constitute one stream of data to be combined with other intelligence streams to inform emergency management decisions. While air sample data are not the sole support for emergency management decisions this study demonstrated that high-volume air sampling equipment can be used to determine the presence and extent of viral agents, supporting emergency responders' risk management decisions.

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