

# Size-specific Filtration Performance of N95 Respirators after Decontamination by Moist Heat Incubation

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## Abstract

### Background

Decontamination and reuse of respirators have been proposed to mitigate the shortage of respirators during pandemics. The US National Institute for Occupational Safety and Health (NIOSH)'s respirator filtration efficiency (FE) test has been used to confirm that decontamination procedures maintain minimum FE above 95% for N95s and similar respirators. However, it was hypothesized that the limited range of test particle sizes may not include the most penetrating particle size for all respirators, especially after decontamination by moist heat incubation (MHI).

### Methods

A custom-designed apparatus was used to measure size-specific FE for respirators across particle size bins between aerodynamic diameter of 0.07  $\mu\text{m}$  and 1.97  $\mu\text{m}$  using an electrical low pressure impactor. FEs were measured for two N95 respirator models before and after ten cycles of MHI. In addition, pressure

drop through respirators and scanning electron microscope (SEM) images of respirator layers were obtained before and after MHI.

## Results

For Kimtech™ brand N95 respirators, FE was not reduced at any size after MHI. For Safe Life brand N95s, FE was below 95% before MHI and decreased significantly after MHI. The most penetrating particle size for this respirator was outside the range defined in NIOSH test protocol, and increased after MHI. There was no appreciable change to the pressure drop through the two respirator models after MHI, nor was any deterioration in fiber integrity visible in SEM images.

## Conclusions

Based on the results of the present study and other studies in the literature, MHI can be used to decontaminate respirators without significant decrease in FE. However, potential effects of MHI on FE need to be assessed for each respirator model. The ability to evaluate size-specific FE across a wide range of particle sizes is important in identifying the most penetrating particle size and associated FE of respirators before and after MHI.

## Introduction

Filtering facepiece respirators (hereafter referred to as respirators) protect against inhalation exposure to particulate contaminants by maintaining a tight seal to users' faces and providing effective filtration over a wide range of particle sizes<sup>1,2</sup>. In North America, certified N95-type respirators with a minimum filtration efficiency (FE) of 95% are widely used for the protection of healthcare workers. These respirators' FEs are tested to be certified by the US National Institute for Occupational Safety and Health (NIOSH) as per procedures listed in Title 42 CFR Part 84<sup>3</sup>. Similarly, the GB 2626-2006 standard is used to certify KN95 respirators in China and the EN 149-2001 standard is used for FFP2 respirators in Europe<sup>4</sup>. The NIOSH test parameters specified in Title 42 CFR Part 84 Subpart K are intended to provide a challenging scenario for

the filtration of small particles and to measure the “worst-case scenario” performance for N-type respirators. These parameters include a high flow rate of  $85\pm 4$  L/min, corresponding to a face velocity of 9.3 cm/s for a typical N95 respirator<sup>5</sup> (flow rate divided by the cross-sectional area of respirator exposed to the flow); use of sodium chloride (NaCl) test particles with a count median diameter of  $0.075\pm 0.20$   $\mu\text{m}$  and a standard geometric deviation not exceeding 1.86, based on reported most penetrating particle sizes (0.030  $\mu\text{m}$  to 0.100  $\mu\text{m}$ ) for N-type respirators<sup>6</sup>; and test particles that are charge-neutralized to achieve Boltzmann equilibrium, giving lower FE compared to charged particles<sup>7</sup>. Before the FE tests, respirators are also preconditioned at 38°C and 85% relative humidity (RH) for 25 hours which may reduce electrostatic deposition<sup>8,9</sup>. The test continues until minimum efficiency is achieved or until a mass of at least  $200\pm 5$  mg of test particles has contacted the filter.

During pandemics caused by infectious viruses such as SARS-CoV-2<sup>1</sup> and H1N1<sup>10</sup> that can transmit through aerosols, demand for respirators has been high. This has led to a shortage of respirators, prompting attempts to increase the supply by decontaminating and reusing available respirators<sup>11</sup>. There is no official decontamination method for respirators but sterilization by vaporous hydrogen peroxide, ultraviolet germicidal irradiation, and moist heat incubation (MHI) are three decontamination methods recommended by the US Centers for Disease Control and Prevention<sup>12</sup> because they have been shown to disinfect respirators with less impact on their filtering performance and fitting than other methods, such as autoclaving and isopropyl alcohol soaking<sup>13</sup>. For certain techniques such as hydrogen peroxide gas plasma<sup>14,15</sup>, multiple cycles of decontamination can lead to decreased protection for users from the loss of FE or deformation of the respirator seal. For N95-type respirators that often utilize electrostatic (electret) media<sup>13</sup>, electrostatic effects may decrease from decontamination methods that require high relative humidity and temperatures or chemical disruption of the fiber – and consequently decrease FE, especially at larger particle sizes up to 0.4  $\mu\text{m}$ <sup>16</sup>. To support and ensure the safety of decontaminated respirators, NIOSH’s FE tests have been used to quantify the FE of decontaminated NIOSH- and non-

NIOSH-certified respirators<sup>10,17,18</sup>. However, the limited range of test particle diameters within the NIOSH test procedure may neglect decreased FE at larger particle diameters and therefore overestimate respirator performance.

We hypothesized that the NIOSH test particle size range may not be sufficient to capture worst-case FE for decontaminated N95 respirators, particularly if a decontamination technique negatively influences electrostatic properties of the respirator material. Moist heat incubation (MHI) was chosen as the decontamination method in the present study, as it has previously been shown effective in deactivating viruses and bacteria while maintaining respirator integrity (Table 1): Heimbuch *et al.* found that MHI fully deactivated H1N1 viruses more consistently than ultraviolet germicidal irradiation<sup>19</sup>. However, previous studies used the limited range of test aerosol sizes defined by NIOSH, or similar, and did not measure particle-size-specific FE. Therefore, we utilized a custom experimental apparatus to extend the range of test particle sizes and to measure FE at different test particle size bins. The FE, pressure drop, and physical integrity of two different, commercially available N95 respirator models labeled as providing at least 95% FE were investigated before and after ten cycles of MHI decontamination.

## Materials and Methods

### Respirator Selection

Two respirator models including the Kimtech™ N95 (53358; Kimberly-Clark Corp., Roswell, GA, USA) and the Safe Life N95 (B130; Safe Life Corp., San Diego, CA, USA) were selected for testing. Two different models were deemed a sufficiently large enough sample group, as the aim of this study was to assess a respirator test methodology evaluating size-specific FE, and not to broadly assess performance of certified versus non-certified respirators. The Kimtech™ N95 53358 is a NIOSH-certified N95 particulate filter respirator (approval number TC-84A-9042) and was acquired through Fisher Scientific in 2020. The Kimtech™ respirators did not have an expiry date indicated but were considered not expired as they were

stored for a maximum of 5 months since their manufacturing date prior to the test: most N95 respirators are likely to maintain the required FE after 10 years.<sup>20</sup> The Safe Life N95 B130 has not been NIOSH-certified since 2015 but it was used in this study as its stockpile was considered for use during the COVID-19 pandemic.<sup>21</sup> The Safe Life respirators were donated from a stockpile of respirators at the University of North Carolina, with a limited sample shipped to Edmonton for inclusion in the present study. The Safe Life respirators did not have the expiry date indicated and were considered near expiration as they were stored for approximately 10 years since their addition to the University of North Carolina's stockpile. For each respirator model, three respirators were randomly selected for the control group and another three for the decontaminated group.

### Moist Heat Incubation Decontamination

Respirators were decontaminated in an environmental chamber (Lunaire CEO910W-4; Thermal Product Solutions, Williamsport, PA, USA) with temperature and humidity control. In each round of decontamination, the decontaminated respirators were conditioned at an average temperature of  $59\pm 1^\circ\text{C}$  and  $67\pm 2\%$  relative humidity (RH) for 30 minutes, then dried at  $59\pm 1^\circ\text{C}$  and  $11\pm 2\%$  RH for another 30 minutes. This was repeated ten times for each sample in the decontaminated group. All respirators were transferred in sealed Ziploc bags at room temperature and tested for FE within 4 days after decontamination.

### Filtration Efficiency Test

Before each FE test, both control and decontaminated respirator samples were preconditioned at  $38^\circ\text{C}$  and 85% RH for 25 hours in the environmental chamber and tested within ten hours, consistent with test procedures outlined in Title 42 CFR Part 84 Subpart K. Previously developed methods<sup>22,23</sup> were modified to measure FE and pressure drop across respirators using the experimental setup detailed in Figure 1. Isotonic saline (0.9% w/v of NaCl) was nebulized with a 6-jet nebulizer (Collison Nebulizer; CH Technologies, Westwood, NJ, USA) using compressed dry air at a pressure of 138 kPa (20 psi). Emitted

droplets were neutralized to a Boltzmann distribution using a Kr-85 charge neutralizer and then dried to solid particles through a silica gel drying column. The test particles, after being well-mixed at the top of a large plenum through two fans, settled into the main chamber of the plenum through a hexagonal mesh serving as a flow straightener. Sample holders on the blank and filter lines were designed such that a 5.72 cm by 8.26 cm (2.25 in by 3.25 in) cutout of respirator material was exposed to the constant 30 L/min flowrate generated by the vacuum pump (the flow rate specification of the electrical low pressure impactor noted below), which corresponds to a face velocity of 10.6 cm/s – similar to the typical NIOSH respirator testing at 9.3 cm/s. Face velocity, rather than flow rate, is the more important factor in determining FE according to single-fiber filtration theory<sup>24</sup>, and other studies also have used different flow rates to achieve face velocities similar to NIOSH's or real-life situations<sup>25,26</sup>. Hence, measurements obtained using the present setup were expected to be comparable to those achieved in a standard respirator test per Title 42 CFR Part 84 for N95 respirators.

Particle concentrations,  $C$ , in each line were measured with an electrical low pressure impactor (ELPI; Dekati Ltd., Kangasala, Finland). Concentrations were averaged over three 100-second periods in the blank line ( $C_{\text{challenge}}$ ) and averaged over two 100-second periods in the filter line ( $C_{\text{filtered}}$ ). The ELPI continuously measured particle concentration as a function of aerodynamic particle size in 12 discrete bins bounded by the aerodynamic diameters ranging from 0.04  $\mu\text{m}$  to 8.15  $\mu\text{m}$ . For this study, only size bins in the range from 0.07  $\mu\text{m}$  to 1.97  $\mu\text{m}$  were used, because particle concentrations outside of this range required over 15% correction on raw measurements to compensate for charger efficiency, bouncing, diffusion, and space charge, and because concentration for particles larger than 1.97  $\mu\text{m}$  was less than 0.1% of total concentration. The chosen range was deemed sufficient as it included NIOSH's standard test particle range (for which 68% of particles have aerodynamic diameters between 0.06  $\mu\text{m}$  to 0.21  $\mu\text{m}$  – as calculated from the nominal count median diameter and geometric standard deviation specified in Title

42 CFR Part 84), but expanded the range to larger particle sizes. The FE in each *i*th bin,  $FE_i$ , was then determined using Equation 1.

$$FE_i = 1 - \frac{C_{\text{filter},i}}{C_{\text{challenge},i}} \quad (1)$$

A manometer was connected between the blank and filter lines to measure pressure drop downstream of the respirator material. Three replicates were performed for each group. Environmental conditions within the plenum during testing were as follows: ambient pressure of  $93 \pm 1$  kPa; temperature of  $23 \pm 1^\circ\text{C}$ ; and relative humidity of  $34 \pm 7\%$  RH.

### SEM Image

Field emission scanning electron microscopy (Zeiss Sigma FE-SEM; Carl Zeiss, Oberkochen, Germany) was used to investigate any possible physical deteriorations in respirator filter layers after decontamination. FE-SEM was operated at electron high tension of 4.00 kV and imaged the filter layers at the magnifications of x40 and x100. The filter layers mounted onto carbon tape were placed over aluminum stubs. Prepared stubs were subsequently coated with gold (Denton Vacuum Desk II Sputter Coater; Denton, Moorestown, NJ, USA) to a thickness of approximately 16 nm.

### Statistical Analysis

Means are expressed with standard deviation over repeated tests as mean $\pm$ SD. Statistical analysis was performed on FE and pressure drop data. For FE, decontaminated respirators were compared with the control group using two-way analysis of variance (ANOVA). For pressure drop, the decontaminated group was compared with the control using two-sample t-test assuming unequal variances. Both analyses assumed a 95% confidence level.

## Results

The size-specific FEs of the two different respirator models before and after ten cycles of MHI are shown in Figure 2, in which the standard NIOSH test particle size range (aerodynamic diameters between 0.06  $\mu\text{m}$  to 0.21  $\mu\text{m}$ ) is indicated as a grey area. The most penetrating particle sizes (MPPS) and corresponding FE are tabulated in Table 2. NIOSH-certified Kimtech™ N95 respirators maintained a FE greater than 98% across the size range of test particles both before and after decontamination cycles. The change in FE between control and MHI groups was 0.4% on average which was not significant ( $p>0.05$ ). The FE of the non-NIOSH-certified Safe Life N95 respirator dropped on average by 6.3% after decontamination, which was statistically significant ( $p<0.05$ ). The MPPS of Safe Life respirators were found to be outside the NIOSH test particle range in both control and decontaminated groups, and the largest drop of 8.4% in FE occurred at 0.49  $\mu\text{m}$ . It was also found that after ten cycles of MHI, Safe Life respirators' MPPS shifted to larger particle size, from 0.32  $\mu\text{m}$  to 0.49  $\mu\text{m}$ .

Ten cycles of MHI did not have appreciable influence on the pressure drop through respirator samples (Table 3). For the Kimtech™ respirator, differences in pressure drop before and after MHI were not statistically significant ( $p>0.05$ ). For the Safe Life respirator, pressure drop decreased after decontamination ( $p=0.04$ ), but the decrease of 12 Pa was deemed negligible from a practical perspective. As shown in SEM images (Figure 3), there were no obvious visible changes to filter layers for any of the respirators.

## Discussion

In the present study, multiple cycles of moist heat incubation did not compromise the FE of the NIOSH-certified N95 respirator tested, the Kimtech™ N95, which agrees with findings in previous studies for different models of certified N95 respirators<sup>10,14,27</sup>. However, this result was not consistent across the two high-efficiency respirator models tested. For the non-NIOSH-certified Safe Life respirators, FE was reduced



across all tested particle sizes after ten cycles of MHI. Furthermore, the most penetrating particle sizes shifted to larger particle sizes, outside the range of those tested in the NIOSH test standard (aerodynamic diameter 0.06  $\mu\text{m}$  to 0.21  $\mu\text{m}$ ). It was initially assumed that the decreased FE would have been mainly caused by either physical deterioration in fibers or decreased electrostatic effect. As SEM images showed that the respirators' fibers were not physically degraded, this decrease in FE and the shift in the most penetrating particle sizes to 0.49  $\mu\text{m}$  may be due to deterioration in electrostatic filtration after exposure to high temperature and humidity. For N95-type respirators in which electrostatic effects play a major role in enhancing FE, the most penetrating particle size tends to lie below 0.10  $\mu\text{m}$ <sup>26</sup>. In contrast, materials that do not have a strong electrostatic effect exhibit most penetrating particle sizes above 0.25  $\mu\text{m}$ , as shown in previous studies in which electrostatic forces have been removed, e.g. by exposure to isopropanol<sup>16</sup>.

It is important to note that respirator certification tests defined by NIOSH and other agencies have been developed to assess protection against a broad range of hazardous aerosols, including dust, fumes, and mists that may be encountered in a workplace setting. Many of these aerosols contain smaller particles than the sizes encountered in the context of protection against infectious aerosols. For instance, the diameter of the SARS-CoV-2 virus is approximately 0.06  $\mu\text{m}$  to 0.14  $\mu\text{m}$ <sup>28</sup> and infectious aerosol particles containing virus must be at least as large as the virus itself, owing to components of lung fluid, saliva, or mucus which will remain even after rapid evaporation of water from exhaled droplets<sup>29</sup>. As a result, the relevant size range for infectious aerosols may exceed the size range used in NIOSH certification testing, and filtration at such sizes cannot be readily inferred from the results of certification testing. The risk associated with neglecting to test filtration for larger particles is greater for infectious aerosols than for many workplace aerosols because larger particles are capable of containing higher viral loads owing to their increased volume. These considerations suggest that for assessing protection against infectious aerosols, respirator FE should be measured at diameters larger than specified in NIOSH test protocol,

especially after decontamination cycles that can reduce electrostatic filtration, and for non-certified respirators or homemade masks<sup>30</sup> that may underperform at larger particles sizes. Reporting FE only within the NIOSH test particle range, or at a specific particle size<sup>31–33</sup>, may not adequately characterize respirator performance against infectious aerosols, and not capture actual worst-case scenarios.

A limitation of the present study is the limited number of samples and the limited pool of respirator types and models tested, as well as the lack of full details of the previous storage conditions, especially for the Safe Life respirators. It is uncertain whether the FE measured for control Safe Life respirators without MHI is reflective of the respirator design itself<sup>20</sup>, the specific samples tested, or whether FE was influenced by storage conditions, for example high humidity<sup>34</sup>. Independent NIOSH test results provided through the US National Personal Protective Technology Laboratory's (NPPTL) beyond-shelf-life and stockpiled respirator assessment indicates that FE for Safe Life B130 respirators which are from three different lot numbers and sourced from at least two different storage facilities, varied from 89.8% to 99.7%.<sup>21</sup> In the present study, FE evaluated for control Safe Life B130 respirators within NIOSH test particle sizes is in the range of the NPPTL test results. However, it should be noted that NIOSH does not have requirements for shelf life or storage conditions for particulate-only air purifying respirators<sup>21</sup> such as N95 hence it is uncertain whether the length of storage and/or storage conditions influenced FE in NPPTL results as well. Due to the lack of records on past history of respirators, the relationship between long term storage conditions and degradation of respirators requires further investigation. Regardless, the present study demonstrates the utility of size-specific FE measurement to identify most penetrating particle sizes, before or after storage, and to evaluate FE at these sizes. Future studies to test size-specific FE of respirators under different MHI temperature or RH conditions and other decontamination methods are also warranted.

Another limitation of the present study was that the same respirator samples were not used for before and after MHI. Measuring FE of a new respirator and then using the same respirator to decontaminate

and measure FE after MHI may have minimized any variations amongst the samples. However, the incremental addition of NaCl aerosol over multiple tests may result in unintended degradation in FE<sup>35</sup>, while this study intended to explore the isolated effect of MHI. Hence, in order to eliminate the possibility that NaCl deposition itself could influence FE measured after decontamination, we chose to use different respirator samples for before and after MHI.

## Conclusion

Two different, commercially available N95 respirator models were decontaminated through ten cycles of MHI, and their FEs were tested in a custom experimental set-up utilizing a range of particle sizes wider than the standard NIOSH respirator certification tests. The encouraging results of the present study for one of the two respirator model studied, coupled with other studies in the literature, suggest that MHI can provide an effective method for decontaminating N95s for reuse. However, for the other respirator model studied, FE was below 95% before MHI cycles, and decreased significantly after MHI cycles. Moreover, the most penetrating particle size for this respirator was outside the range defined in NIOSH certification testing, and increased after MHI cycles. The ability to evaluate size-specific FE across a wide range of particle sizes, as presented herein, is important in identifying the most penetrating particle size and associated FE of respirators.

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## Authorship Confirmation Statement

Design of experiment set-up: C.A.R, H.W, W.H.F, A.R.M. Filtration efficiency test: B.J., S.S. Data analysis and interpretation: S.S. All authors contributed to the revision of the article.

## Authors' Disclosure Statement

The authors declare they have no competing financial interests.

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Table 1

Comparisons between current and previous studies on the effects of moist heat incubation (MHI) on respirators

	Current Study	Heimbuch et al. (2011) <sup>19</sup>	Bergman et al. (2011) <sup>10</sup>	Daeschler et al. (2020) <sup>18</sup>	
Respirator Tested	<ul style="list-style-type: none"> <li>• Kimtech™ N95 53358</li> <li>• Medstar KN95</li> <li>• Safe Life N95 B130</li> </ul>	<ul style="list-style-type: none"> <li>• Three NIOSH- and FDA-approved N95 surgical respirators</li> <li>• Three NIOSH-approved N95 particulate respirators (Makes and brand names anonymized)</li> </ul>	<ul style="list-style-type: none"> <li>• 3M 1860</li> <li>• 3M 1870</li> <li>• Kimberly Clark PFR95-270 (46767)</li> </ul>	<ul style="list-style-type: none"> <li>• 3M 1860</li> <li>• 3M 8110</li> <li>• 3M 8210</li> <li>• 3M 9105</li> </ul>	
Number of Decontamination Cycles	10	1	3	10	
Heating Condition	59±1°C and 67±2% RH for 30 minutes	65°C±5°C and 85±5% RH for 30 minutes	60°C and 80% RH for 15 minutes	70°C and 50% RH for 60 minutes	70°C and 0% RH for 60 minutes*
Drying/Cooling Condition	59±1°C and 11±2% RH for 30 minutes	Not described	Not described	Room temperature for 5 minutes mid-cycle	Room temperature for 5 minutes mid-cycle
Range of NaCl Test Particle Used	Wider than NIOSH's range: aerodynamic diameter 0.07 to 1.97 µm (approx. count diameter 0.05 to 1.34 µm)	Not tested against test particles	Same as NIOSH's range: count median diameter of 0.075 ± 0.020 µm and a geometric standard deviation of less than 1.86	Similar to NIOSH's range: count median diameter of 0.075 ± 0.020 µm	
Physical Degradation	See Results	No obvious visual deterioration	Mean face seal leakage <1% throughout the three cycles of decontamination	Minimal change in fiber diameter No drop in filtration efficiency	
Microbial Inactivation	Not examined	>4 log reduction of H1N1	Not examined	Density of <i>E. coli</i> decreased from 2.77 to 0.02 through optical density measurement	No infectious SARS-CoV-2 detected

\*Note that this dry heat condition (0% RH) is not an example of moist heat decontamination, however, it demonstrated that SARS-CoV-2 are susceptible to destruction under high temperature. This implies that it will be more susceptible to destruction under high temperature accompanied by high humidity as moist heat can destroy proteins more efficiently than dry heat<sup>20</sup>.

Table 2

Filtration efficiency (FE) at the most penetrating particle size (MPPS) before and after ten cycles of moist heat incubation (MHI)

		Kimtech™ N95	Safe Life N95
Control	FE	98.4±0.7%	89.4±2.0%
	MPPS	0.12 µm	0.32 µm
After Ten Cycles of MHI	FE	98.2±0.0%	81.1±3.1%
	MPPS	0.12 µm	0.49 µm

Table 3

Pressure drop across commercial N95s before and after ten cycles of moist heat incubation (MHI)

	Kimtech™ N95	Safe Life N95
Control	106±4 Pa	64±4 Pa
After Ten Cycles of MHI	109±1 Pa	52±1 Pa

## Figure 1

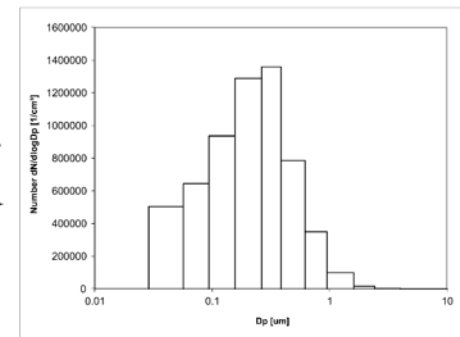
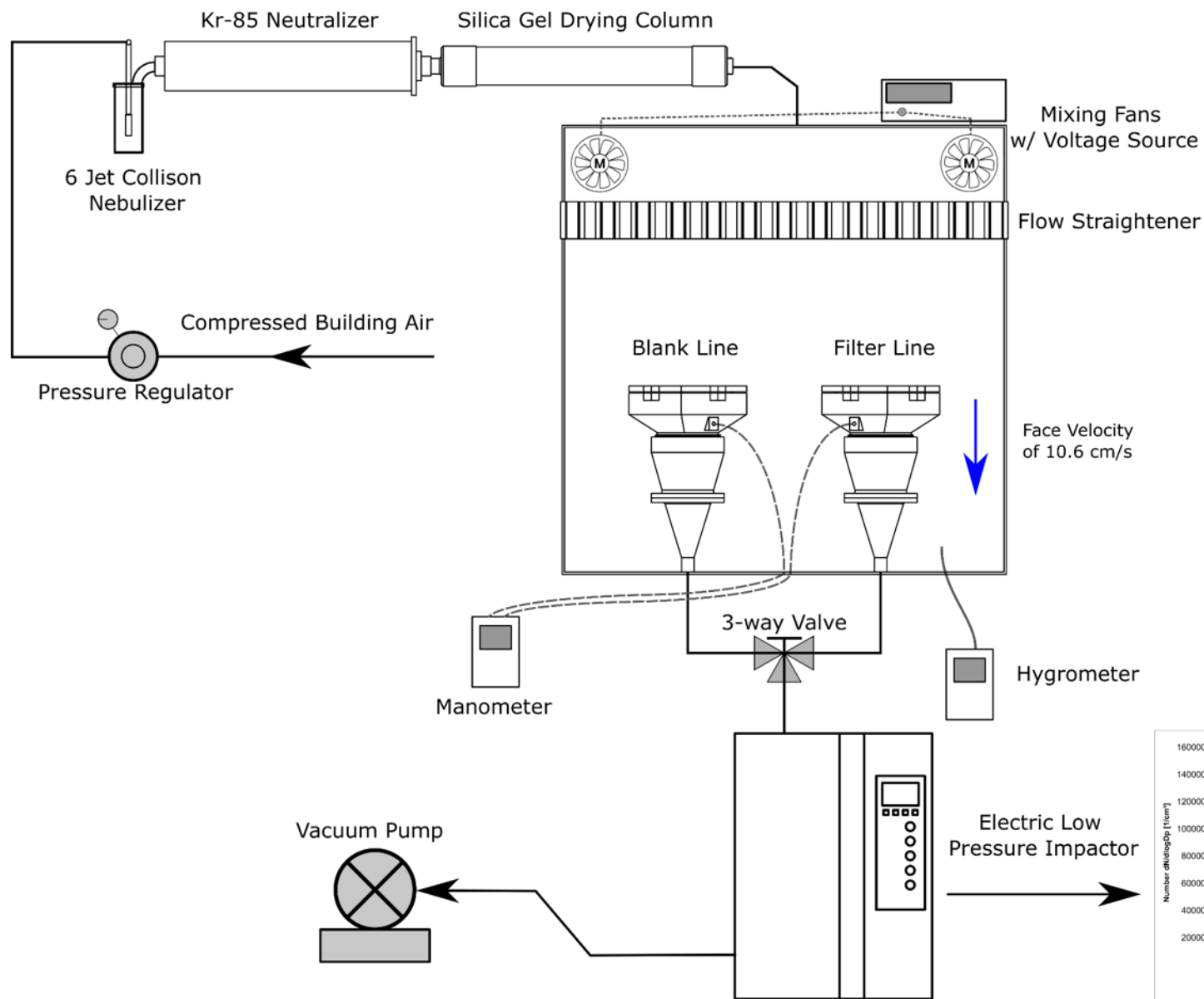
Experimental setup used to quantify filtration efficiencies across a range of particle sizes and pressure drops. A comparison of number concentrations measured by Electric Low Pressure Impactor in the blank and filter lines drawing from well-mixed test particles in a large plenum allowed quantification of filtration efficiencies across various particle sizes. (Adapted from Phillip Clapp et al.: A Simple Homemade HEPA Filtering Facepiece. Emerg Infect Dis. Manuscript in review).

## Figure 2

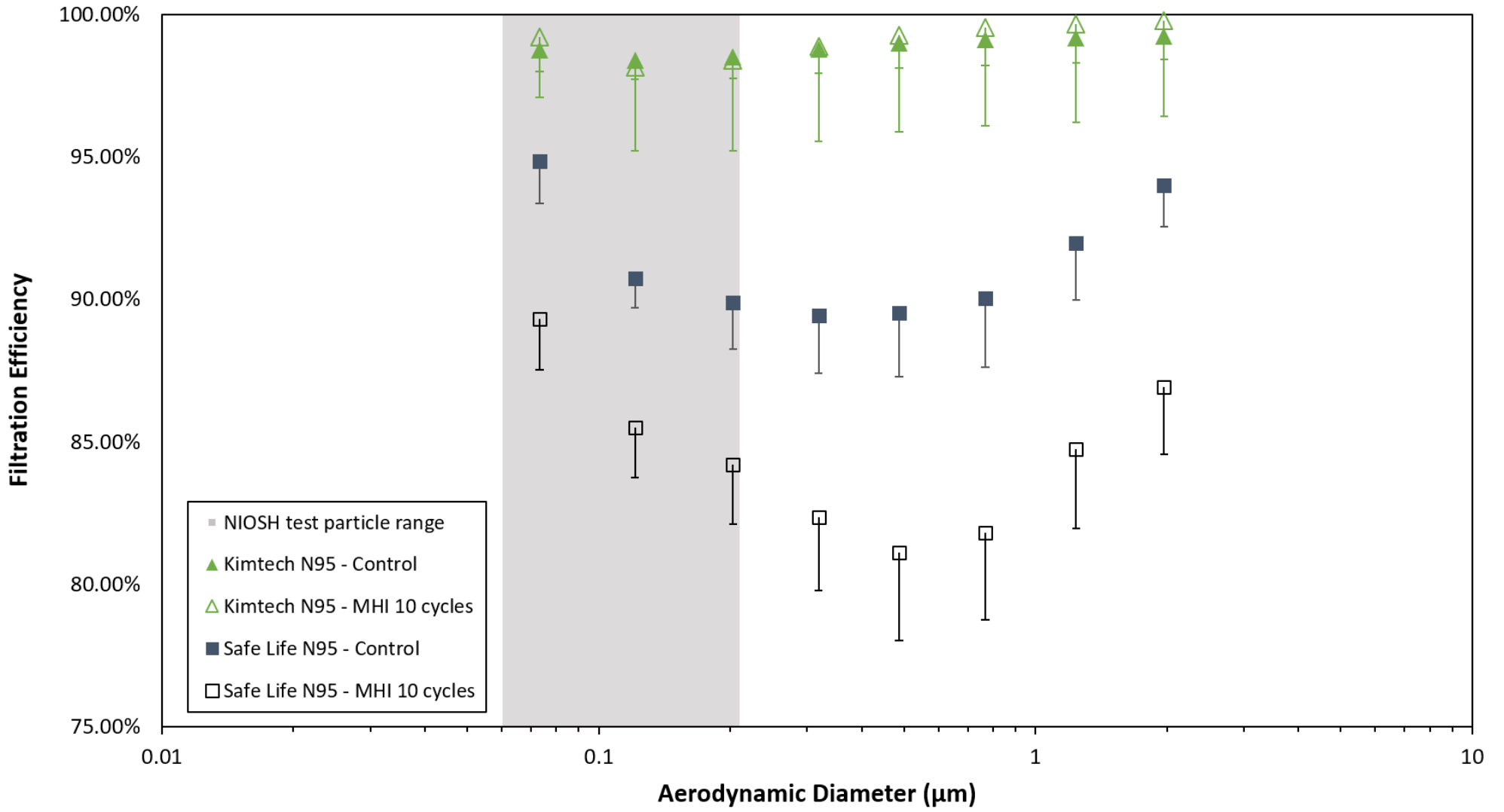
Filtration efficiencies from commercial N95s before and after ten cycles of moist heat incubation (MHI). Grey area indicates typical NIOSH test particle sizes (aerodynamic diameter of 0.06  $\mu\text{m}$  to 0.21  $\mu\text{m}$ ). For clarity, only negative error bars are depicted.

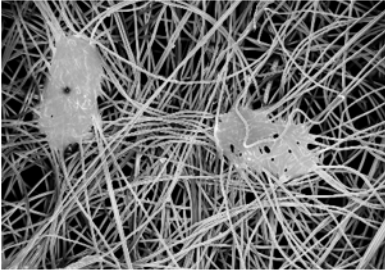
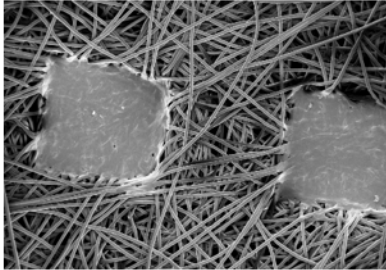
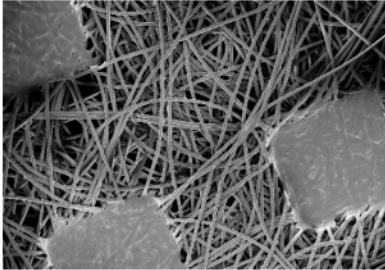
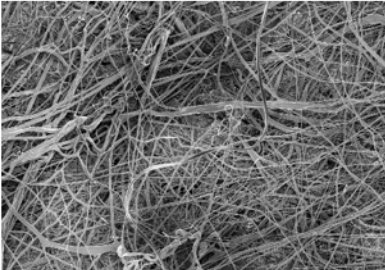
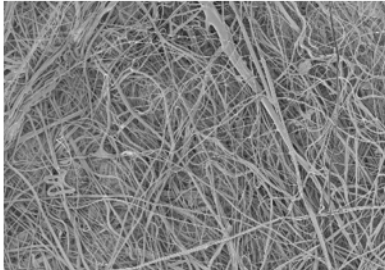
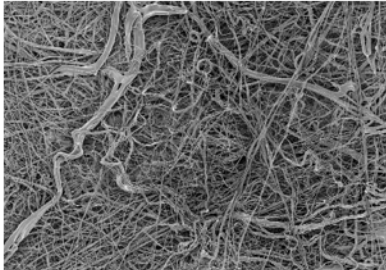
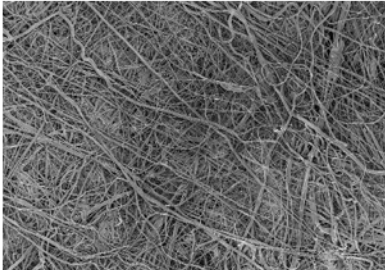
## Figure 3

SEM images of commercial N95 respirators before and after ten cycles of moist heat incubation (MHI).



Particle Concentrations  
 from 0.07 to 1.97  $\mu\text{m}$   
 Over 8 Bins



	Kimtech™ N95		Safe Life N95	
	Control	After Ten Cycles of MHI	Control	After Ten Cycles of MHI
<b>Outer-most Layer</b> (Mag 40x) 500 μm ↔				
<b>Middle Layer</b> (Mag 100x) 100 μm ↔				
<b>Inner-most Layer</b> (Mag 40x) 500 μm ↔	