

Original Article

Quantitative Fit Evaluation of N95 Filtering Facepiece Respirators and Coronavirus Inactivation Following Heat Treatment

Travis L. Massey^{*,}, Monica K. Borucki, Samuel Y. Paik, Kyle W. Fuhrer, Mihail Bora, Staci R. Kane, Razi-ul M. Haque and Salmaan H. Baxamusa

Lawrence Livermore National Laboratory, 7000 East Avenue, Livermore, CA 94550, USA

*Author to whom correspondence should be addressed. Tel: +1-925-422-9509; e-mail: massey21@llnl.gov

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Abstract

Reuse of filtering facepiece respirators (FFRs, commonly referred to as N95s) normally meant for single use has become common in healthcare facilities due to shortages caused by the COVID-19 pandemic. Here, we report that murine hepatitis coronavirus initially seeded on FFR filter material is inactivated (6 order of magnitude reduction as measured by median tissue culture infective dose, TCID₅₀) after dry heating at 75°C for 30 min. We also find that the quantitative fit of FFRs after heat treatment at this temperature, under dry conditions or at 90% relative humidity, is not affected by single or 10 heating cycles. Previous studies have reported that the filtration efficiency of FFRs is not negatively impacted by these heating conditions. These results suggest that thermal inactivation of coronaviruses is a potentially rapid and widely deployable method to reuse N95 FFRs in emergency situations where reusing FFRs is a necessity and broad-spectrum sterilization is unavailable. However, we also observe that a radiative heat source (e.g. an exposed heating element) results in rapid qualitative degradation of the FFR. Finally, we discuss differences in the results reported here and other recent studies investigating heat as a means to recycle FFRs. These differences suggest that while our repeated decontamination cycles do not affect FFR fit, overall wear time and the number of donning/doffing cycles are important factors that likely degrade FFR fit and must be investigated further.

Keywords: coronavirus; decontamination; filtering facepiece respirator; fit; heat; mask; N95; reuse

Introduction

The worldwide global demand for N95 filtering facepiece respirators (FFRs) for healthcare professionals quickly outpaced supply during the global COVID-19

pandemic. Several protocols for disinfecting and reusing FFRs that are normally for one-time use only have been proposed (CDC, 2020), and the FDA has authorized emergency use of vaporized hydrogen peroxide (VHP)

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What's important about this paper

Shortages of single-use filtering facepiece respirators (FFRs, commonly referred to as N95s) are widespread due to the COVID-19 pandemic, leading to an evaluation of reuse options. Thermal inactivation of coronaviruses is a potentially rapid and widely deployable decontamination method to reuse N95 FFRs in emergency situations that does not compromise form, fit, or filtration performance.

as a broad-spectrum sterilant for reuse of FFRs (Hinton, 2020a,b). Because VHP requires centralized operations and specialized equipment, it may not be available in all emergency situations.

Heat treatment, though not a broad-spectrum sterilant, may be easily adapted in a variety of settings as a potential emergency treatment method for recycling N95 FFRs. Filter efficiency of FFRs has been reported to be maintained after dry heat treatment at 90°C for 1 h (Viscusi *et al.*, 2009). A more recent report found that filter efficiency of melt-blown polypropylene filter fabric was not affected by either dry or moist heat at $\leq 85^{\circ}$ C for up to 50 cycles (Liao *et al.*, 2020).

In addition to efficiently capturing aerosolized particles in the filter elements, the FFR must also form a good seal around the nose and face of the wearer; this is fundamental to the overall performance of the FFR. Reports are now emerging on the quality of the seal via quantitative fit testing of FFRs following decontamination protocols. A recent evaluation of typical hospital decontamination protocols showed that quantitative fit was retained following multiple cycles of ethylene oxide, VHP, and (for some FFR models) autoclaving (Kumar et al., 2020). Another recent study, released during the preparation of the present report, reported that the quantitative fit factor of FFRs is retained for two cycles of dry heat at 70°C (duration not stated) but failed thereafter (Fischer et al., 2020). These decontamination cycles were interleaved with participants wearing the FFRs, however; the study does not distinguish whether the failure was due to the decontamination procedure, the wear procedure, or the interplay of the two.

Although heating at these temperatures is not a broad-spectrum sterilant, previous studies in liquid media have reported SARS-CoV-1 inactivation at 60°C for 30 min (\geq 5 log (10^{\geq 5}) reduction in viral activity; Rabenau *et al.*, 2005) and at 75°C for 15 min (\geq 4 log reduction; Darnell *et al.*, 2004). Most recently, SARS-CoV-2 has been shown to be inactivated (\geq 6.8 log reduction) by heating to 70°C for 5 min in liquid media (Chin *et al.*, 2020). While promising, confirmation that the virus can be inactivated after deposition on FFR

filter material is needed to investigate FFR reuse. A study released during the preparation of the present report found that SARS-CoV-2 on FFR filter material could be inactivated (\geq 4 log reduction) after dry heating at 70°C for 60 min (Fischer *et al.*, 2020). Campos *et al.* (2020) likewise demonstrated 4–5 log reduction at 75°C and above, but only 2–3 log reduction at 60°C; humidity was positively correlated with viral inactivation at both temperatures.

Comparison of SARS-CoV-2 stability as a function of temperature on other surfaces can be challenging, as noted by Corpet (2021), because the moisture and wetting properties of the surface play a significant role. Furthermore, when heated, the thermal characteristics of the substrate materials and heating system must also be considered-such as thermal mass, thermal conductivity, mechanism of heat transfer, infrared absorbance, emissivity, etc. For example, Fischer et al. (2020) found that SARS-CoV-2 remained active longer on steel than polypropylene when heated, while Corpet (2021) reported greater viral stability on polypropylene than steel under room temperature conditions. This highlights that while reports of viral stability as a function of surface, temperature, humidity, and time, and experimental configuration may serve as valuable starting points, viral stability should be evaluated under the specific conditions of each use case.

This study first examines whether the quantitative fit of N95 FFRs is impacted by single or repeated heat treatments (dry and humid), then determines whether the dry heat treatment protocol presented is sufficient to inactivate a murine hepatitis virus (MHV) as a surrogate for SARS-CoV-2. We opted to test 75°C cycles, as this provides a 10°C margin from the 85°C limit above which filter damage may occur while remaining sufficiently above the minimum inactivation temperatures. Likewise, the prior literature suggest that 30-min cycles are sufficient but not excessive; rapid decontamination cycles may be important in emergency situations. In addition, not donning the FFRs between decontamination cycles allows us to isolate the effects of the heat treatment from the confounding effects of donning, doffing, and wear time on the quantitative fit.

Methods

Quantitative fit testing

Prior to heat treatment, each volunteer participant briefly fitted a new, unused FFR to his face and nose structure to simulate a first-time use. Volunteers were trained on the use of FFRs by the Respirator Services staff at the Lawrence Livermore National Laboratory (LLNL) and were given donning and doffing instructions. 3M Model 8210 N95s (3M, St. Paul, MN) were used for all the heat treatment tests in this study. This particular model of N95 is one of the most widely recognizable and used N95 FFRs in the industry and is available in one size as it was designed to seal effectively against most human faces. Since a goal of this study was to determine how fit from an initially well-fitting N95, as quantified by fit testing, would be affected by single or multiple heat treatment cycles, and not how different N95 models, at different sizes, would perform for a variety of face structures (other studies have investigated this), only one model of N95 and two volunteer participants were used for the tests in this study. After the initial donning/doffing cycle and prior to heating, the FFRs were loaded into sterilization pouches (CrossTex Sure-Check, SCL12182). Two FFRs were loaded into each pouch.

Quantitative fit tests were performed per Occupational Safety and Health Administration (OSHA) fit test protocol 1910.134, Appendix A (OSHA, 1998) using a TSI PortaCount Respirator Fit Tester 8038 (TSI Instruments, Shoreview, MN). This instrument does not test the effectiveness of the filter, which previous studies have validated up to 90°C (Viscusi *et al.*, 2009; Liao *et al.*, 2020). The tests performed in this study quantify changes to the sealing surfaces insofar as they affect fit. The OSHA passing criterion for half mask respirators, including FFRs, is a quantitative fit factor of 100. The instrument calculates fit factor as the ratio of aerosolized particle counts per unit volume outside the FFR to inside.

A sodium chloride (NaCl) aerosol generator and two humidifiers with tunable droplet size set at the smallest droplet size setting were used to achieve background levels of aerosol. The PortaCount Fit Tester 8038 uses a preselector to ensure that the detected aerosols reflect those penetrating the sealing surfaces and not the filter itself. Aerosolized particle counts were compared inside and outside the FFR while the participant performed a series of seven 60-s and one 15-s breathing, movement, and speaking exercises including:

- Normal breathing
- Deep breathing
- Head side-to-side
- Head up and down

- Talking
- Grimacing (15 s)
- Bending over/reaching down
- Normal breathing

Samples were fit tested on the same volunteer who donned and doffed the FFR prior to the heat treatment. Fit tests on samples 01-08 (Volunteer A) were performed sequentially, but the order of fit tests on samples 09-16 (Volunteer B) was randomized. Menton–sellion length and bizygomatic breadth were measured for both volunteers, following the procedure of Bradtmiller and Friess (2004).

A total of 18 FFRs were tested in this study as shown in Table 1. For each heating schedule tested, the samples were cycled either once or 10 times. Because the fit test requires inserting a metal rivet into the FFR, the test was considered destructive (see Discussion). Therefore, preand post-treatment measurements were not possible on the same FFR; instead, control measurements were made for each volunteer with new, unused FFRs without heat treatment.

Dry heating

For dry heating, FFRs (samples 01-08) were loaded into a laboratory oven (Cascade Tek TFO-1). One pouch of two FFRs was placed on each of two shelves within the oven. The oven was prewarmed to 75°C and operating at ambient humidity. We estimate that humidity at 75°C was approximately 2.5% relative humidity, based on a measured 40% ambient relative humidity at 20°C in the room, no added water vapor, and a 16.5 times greater moisture capacity in air at 75°C compared with 20°C (Engineering ToolBox, 2009). Ambient temperature and humidity were measured using a commercial integrated thermometer and hygrometer with an attached probe (VWR Traceable Excursion-Trac, model 6452), but the oven temperature exceeded the operating range of the probe and a direct humidity measurement was not possible. The oven door was held open for <30 s during loading and experienced a temperature loss of <2°C.

 Table 1. Sample set of FFRs and conditions tested in this study.

FFR sample	Humidity	Heating cycles	Volunteer	
01, 02, 03, 04	Ambient (<5% RH)	1×	А	
05, 06, 07, 08	Ambient (<5% RH)	10×	А	
17 (control)	N/A	None	А	
09, 10, 11, 12	90% RH	1×	В	
13, 14, 15, 16	90% RH	10×	В	
18 (control)	N/A	None	В	

Some FFRs were instrumented with a thermocouple as shown in Fig. 1a to monitor the thermal history. Fig. 1b shows the thermal history of several FFRs from both the top and bottom shelf of the oven. The FFRs heated to 75° C in approximately 5 min and this temperature was maintained to $\pm 3^{\circ}$ C throughout the 30-min treatment. FFRs that were subjected to multiple cycles were allowed to cool to room temperature but were not removed from the pouch or redonned prior to the next heating cycle.

Humid heating

For humid heating, FFRs (samples 09-16) were loaded into a benchtop environmental chamber (Espec SH-242) in groups of four (two pouches of two FFRs each, as described previously), as shown in Fig. 1c,d. Care was taken not to crush or deform the FFRs. The chamber, which is designed to ramp to and hold a set of programmed temperature and humidity profiles over time, was programmed to ramp over 15 min to 75°C and 90% relative humidity, hold those conditions for 30 min, and ramp back down to room temperature and humidity over 15 min. For FFRs treated for multiple cycles, these room temperature conditions were held for 7 h before the next heat and humidity cycle began. We visually confirmed that the pouch allowed steam to permeate to the surface of the FFR during experimental process development. The FFRs were not removed from the sterilization pouch or redonned between cycles.

Neither the oven (dry heating) nor environmental chamber (humid heating) had exposed heating elements, as we found during preliminary tests that the infrared radiation emitted from exposed heating elements was absorbed by the polymer components of the FFR and caused rapid heating and damage to the FFR (see Discussion).

Thermocouple data analysis

Thermocouple (type K) temperature data were logged at 0.6 samples per second per thermocouple. Data were streamed in real time from an Arduino to a Windows computer during dry heating cycles and later plotted in MATLAB. In order to meaningfully overlay temperature plots over multiple cycles, plotting began when the temperature first exceeded 23°C on *either* the top or bottom shelf of the oven, and plotting ended when *both* thermocouples dropped below 23°C. The data were not otherwise processed or subsampled.

Viral activity measurements

MHV (a mouse coronavirus) was used as a surrogate for SARS-CoV-2. MHV, like SARS-CoV-2, is of the genus *Betacoronavirus* and has similar thermal inactivation

kinetics (Guillier et al., 2020). Twenty microliters of viral stock diluted in Dulbecco's modified Eagle medium (DMEM) media (Thermo Scientific, Waltham, MA) with 10% fetal bovine serum (FBS) and antibiotics was inoculated onto 12 replicate 5 mm square coupons cut from an FFR (3M Model 8210). The inocula were dried inside sterile Petri plates within a biosafety cabinet for 1 h, and two sets of three coupons were placed in two autoclave bags (CrossTex Sure-Check, SCL12182), sealed, and placed in the oven preheated to 75°C (Cascade Tek TFO-1). At 30 min one bag containing three coupons was removed from the oven and allowed to cool for 30 min, and the second bag containing three coupons was removed from the oven at 60 min and allowed to cool for 30 min. Control coupons were stored at room temperature and were processed at the same timepoints as when the corresponding 30- and 60-min coupons were processed. Thirty minutes after removal from the oven, each coupon was immersed in 2 ml of media (DMEM, 10% FBS, and antibiotics) and vortexed intermittently for 10 min to dislodge the viral particles from the coupon into the media. The virus was then titered by TCID₅₀ assays using 17CL-1 murine cells. TCID₅₀ is a measure of the concentration of *infectious* virus particles in a sample (counts per volume) determined by the proportion of cell cultures infected at each of several dilutions. Cytopathic effect for each well was recorded on day 3 post-inoculation, and TCID₅₀ titer was calculated using the Spearman and Karber method (Hierholzer and Killington, 1996). TCID₅₀ is plotted with a \log_{10} -scale concentration axis, and reductions in TCID₅₀ are reported as 'N log'-that is, N orders of magnitude, or a factor of 10^N .

Results

Fit test volunteer facial measurements

The facial measurements of the two volunteers and the median US adult (18–66 years; both sexes) are provided below with approximate percentiles relative to the US adult population (Bradtmiller and Friess, 2004).

Volunteer A (male, age 33): Menton–sellion length: 121 mm (60th percentile) Bizygomatic breadth: 138 mm (40th percentile)

Volunteer B (male, age 37): Menton–sellion length: 106 mm (5th percentile) Bizygomatic breadth: 121 mm (1st percentile)

Median US adult and standard deviation: Menton–sellion length: 119.3 ± 8.1 mm Bizygomatic breadth: 140.5 ± 7.9 mm



Figure 1. (a) Thermocouple attached to FFR with Kapton tape. Photograph shows a 3M Model 8511 N95 FFR used for validating the process. All fit tests were performed with a 3M Model 8210 N95 FFR. (b) Thermal history of FFRs on top (light gray) and bottom (dark gray) shelves of oven over 10 cycles. (c) Representative example of four FFRs loaded into pouches and (d) in the Espec SH-242 environmental chamber.

Quantitative fit test results

Results of the quantitative fit tests are shown in Table 2. A passing score is 100, and the maximum score reported by the TSI PortaCount 8038 is 200.

None of the processed samples showed any qualitative change in feel or appearance; however, one of the elastic straps on sample 05 snapped upon doffing the FFR after passing the quantitative fit test.

All samples subjected to dry heat cycles passed the quantitative fit tests with a fit factor of >100. After

one heating cycle there was no significant change to the quantitative fit test result for the four tested samples. After 10 cycles, all 4 tested samples had the maximum score upon fit testing. Due to FFRs 01-08 being tested sequentially, we are unable to disambiguate whether the improvement in quantitative fit test results for FFRs treated for 10 cycles versus one cycle is due to the heat treatment, an improvement in donning procedure over time, or merely coincidental.

FFR sample	Humidity	Heating cycles	Volunteer	Fit factor
01	Ambient (<5% RH)	1×	A	107 (pass)
02	Ambient (<5% RH)	1×	А	195 (pass)
03	Ambient (<5% RH)	1×	А	165 (pass)
04	Ambient (<5% RH)	1×	А	149 (pass)
05	Ambient (<5% RH)	10×	А	200+ (pass) ^a
06	Ambient (<5% RH)	10×	А	200+ (pass)
07	Ambient (<5% RH)	10×	А	200+ (pass)
08	Ambient (<5% RH)	10×	А	200+ (pass)
17 (control)	N/A	None	А	181 (pass)
09	90% RH	1×	В	200+ (pass)
10	90% RH	1×	В	163 (pass)
11	90% RH	1×	В	200+ (pass)
12	90% RH	1×	В	200+ (pass)
13	90% RH	10×	В	200+ (pass)
14	90% RH	10×	В	200+ (pass)
15	90% RH	10×	В	200+ (pass)
16	90% RH	10×	В	161 (pass)
18 (control)	N/A	None	В	200+ (pass)
18 (control)	N/A	None	В	200+ (pa

Table 2. Results of quantitative fit tests.

^aStrap broke upon doffing.

Likewise, all samples subjected to moist heat cycles passed the quantitative fit tests with a fit factor >100. There was no significant change to the quantitative fit test result for the eight samples tested, nor a correlation between score and number of treatment cycles. No significant difference was observed between the two volunteers.

Viral activity results

Dry heat inactivation of MHV, a coronavirus previously used as a SARS-CoV-1 surrogate virus for validating decontamination protocols (Casanova et al., 2010), was used to confirm that heating to 75°C for 30 min inactivates high titers of coronavirus on FFR filter material. Diluting MHV in media containing 10% FBS prior to inoculating the FFR filter material simulated the presence of proteins found in respiratory secretions that may increase viral resistance to inactivation (Casanova et al., 2010). No viral activity was detected after the heat treatment. TCID₅₀ measurements on the coupons used in these studies show $\geq 6 \log$ reduction after heating to 75°C for either 30 or 60 min as compared with room temperature activity for similar time periods as seen in Fig. 2. These results are generally consistent with a recent report that 70°C dry heat for 60 min inactivates SARS-CoV-2 on FFR filter material (\geq 4 log reduction; Fischer et al., 2020) and provide further evidence that dry heat provides relatively rapid inactivation of coronaviruses on FFR filter material, especially as compared



Figure 2. Viral activity after 30 or 60 min of dry heat (75°C) and after 30 or 60 min at room temperature (RT). Heated samples showed a \geq 6 log reduction in viral activity.

with untreated FFRs stored for the same amount of time at room temperature.

Discussion

Selection of subjects and FFRs

We evaluated only one model of FFR due to the limited availability of FFRs at the time of the study. Twenty 3M 8210 FFRs were generously provided by Lawrence Livermore National Laboratory's Health Services Department. Sixteen FFRs (four replicates for four experimental conditions) were used for heat treatment prior to fit testing. Two FFRs were reserved for quantitative fit test control samples. The remaining two FFRs were initially held as additional control samples in case either subject failed to pass the control fit test additional fit test and volunteers needed to be recruited. These two FFRs were later used for viral inactivation testing (see Viral activity measurements). We were limited to two subjects based on both the number of FFRs available for testing and personnel available during a period of strict COVID-19 restrictions. These both represent limitations of the study, but do not, in the authors' opinions, alter or significantly weaken the conclusions of the quantitative fit testing portion of the study.

Because quantitative fit testing is a destructive process, the two fit test control FFRs also served as pretreatment baseline measurements for each subject. The metal port required for quantitative fit testing has a high thermal conductivity relative to the melt-blown polypropylene filter and a low thermal mass, so we could not rule out that the presence of this metal port could alter the heat concentration and the results of the post-heattreatment quantitative fit tests. As such, quantitative fit tests were considered destructive, and these FFRs were not reused for subsequent heat treatment cycles and quantitative fit tests. This concern could be addressed in a future study in which there are ample FFRs to evaluate whether the metal port being present during heat treatment alters the results of the post-treatment quantitative fit test.

Selection of heating method may impact fit, function, and safety

In preliminary experiments not reported in this study, we screened several methods of heating unused N95 FFRs using household appliances as an alternative to laboratory or clinical ovens. In one test, we heated an FFR in an oven (set to 75°C) with an exposed heating element. The FFR heated rapidly and showed visible signs of softening and melting. We believe this is due to the infrared radiation emitted by heating elements, which typically operate at temperatures of approximately 800–1000°C. Polymers strongly absorb blackbody radiation (3–4 µm wavelength) emitted by the heating elements at this temperature (Brochocka *et al.*, 2020). We therefore caution against using any heating method which exposes the FFR directly to radiation from the heat source. We also briefly considered heating of N95 FFRs in a residential clothes dryer as a rapidly deployable solution, but temperature was poorly controlled and not repeatable across runs on one dryer. Even if an industrial dryer's heating profile could be adequately characterized and controlled, there was concern over poorly sealed dryer doors or ductwork facilitating spread of aerosolized virus. As such, this method of heating was rejected.

Because the fit test results and viral inactivation have only been demonstrated under laboratory heating conditions, we strongly advise against generalizing this procedure to poorly controlled do-it-yourself solutions until many studies' aggregate fit test and coronavirus inactivation results can better inform the functional bounds of a heat-based decontamination procedure. Each implementation of a heat-based decontamination system following a similar protocol should be validated prior to deployment.

Donning, wear time, and doffing likely degrade FFR fit

While we found that the quantitative fit factor of FFRs was not affected by up to 10 30-min heating cycles at 75°C, a recent report by Fischer et al. (2020) found that the quantitative fit factor of FFRs was only retained for two cycles of 70°C heat treatment (they tested up to three cycles). The key difference in the two studies was the treatment of FFRs between heating cycles. In the present study, we donned and doffed the FFR a single time but did not simulate donning and doffing in between heating cycles-an important limitation of our study. In Fischer et al. (2020), the FFR was donned and worn for 2 h between cycles. Because this study clearly establishes that similar heat treatment does not compromise the fit of the FFR, Fischer's results suggest that use duration and number of donning/doffing cycles of the FFR, perhaps even independent of heat treatment (or other decontamination protocol such as VHP), likely play an important role in the quantitative fit factor of FFRs that are utilized beyond the recommended single use. Bergman et al. (2012) likewise demonstrated that repeated donning and doffing cycles degraded the fit of six FFR models, but the mechanism of degradation was not identified.

Additional studies are needed to disambiguate the effects of total use duration, donning/doffing cycles, and decontamination protocol on the quantitative fit factor of FFRs and in particular on the elastic head and neck straps. Wearing the FFR represents a continuous low-level strain on the elastic bands, and donning and doffing induces a large strain for a short period of time. Certain decontamination procedures, such as ultraviolet irradiation, extreme heat, or a strong oxidant, could embrittle or otherwise degrade the elastic bands; thus, wear time and donning/doffing cycles should be evaluated in conjunction with the decontamination procedure.

Conclusion

We subjected N95 FFRs to 1 and 10 heating cycles up to 75°C under dry and humid (90% relative humidity) conditions. Quantitative fit testing did not show any degradation in the fit factor, indicating that the form and fit of these FFRs was retained following the heat treatment. There was no measurable difference in quantitative fit between the dry or humid heating protocols for the temperature, duration, and humidity levels tested. We also found that dry heating to 75°C reduced the viral activity of MHV on an FFR filter element by $\geq 6 \log$. These temperatures and times have already been shown to inactivate other coronaviruses (including SARS-CoV-2) in liquid media (Darnell et al., 2004; Rabenau et al., 2005; Chin et al., 2020) and FFR filter material (Fischer et al., 2020), and our study provides further evidence that virus dried on FFR filter material can also be inactivated by heating. Previous studies have shown that these temperatures do not negatively impact filter efficiency and airflow of melt-blown propylene filter elements found in N95 FFRs (Viscusi et al., 2009; Liao et al., 2020).

The emerging evidence supports that heat treatment may be used as an effective method for reusing N95 FFRs. It should be noted that heat treatment is not a broad-spectrum sterilant and that N95 FFRs are normally meant for one-time use. However, in emergency situations heat treatment protocols specifically to inactivate coronaviruses may be developed using commonly available equipment (incubators, blanket warmers, ovens, etc.). Heat treatment may therefore serve as a rapid method for reuse of FFRs in areas where FFRs are in critically short supply, specialized decontamination equipment (e.g. VHP) is not available, and surface sterilization (e.g. ultraviolet germicidal irradiation) is insufficient. However, important questions remain on the retention of fit factor after long-term use and repeated donning/doffing cycles to help resolve conflicting data on the number of cycles for which quantitative fit factor can be maintained.

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Conflict of interest

The authors declare no conflict of interest relating to the material presented in this article. Its contents, including any opinions and/or conclusions expressed, are solely those of the authors. This document is for technical information purposes only and is not a substitute for independent medical judgment.

Data availability

The data underlying this article will be shared on reasonable request to the corresponding author.

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