A Scalable Method for Ultraviolet C Disinfection of Surgical Facemasks Type IIR and Filtering Facepiece Particle Respirators 1 and 2

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Abstract

Due to the SARS-CoV-2 pandemic a shortage of personal protective equipment, including surgical facemasks and Filtering Facepiece Particle Respirators has occurred. SARS-CoV-2 has a 79.5-82% homology to SARS-CoV. The SARS-CoV UVC sensitivity is described in literature. We have performed UVC transmission measurements of surgical facemasks and respirators. In addition, we performed UVC disinfection experiments of *S. aureus* with surgical facemasks and respirators. Results show that we can achieve an 8-log reduction of *S. aureus* in the inner layers of FFP1 respirators and the exterior of surgical facemasks. Furthermore, we showed a 7-log reduction of *S. aureus* in the inner layers of FFP2 respirators. We conclude that UVC disinfection is an effective, safe and scalable method for reuse of surgical facemask and respirators.

Keywords: SARS-CoV2; UVC; disinfection; respirators; reuse

Introduction

Coronavirus disease 2019 (COVID-19) is caused by Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2)\(^1\). It is current understanding that SARS-CoV-2 is transmitted between persons via respiratory droplets (>5-10 µm) and fomites. There is no evidence for airborne transmission via aerosols or droplet nuclei (<5 µm)\(^2\-5\). There are, however, medical procedures which produce aerosols e.g.: endotracheal intubation, bronchoscopy, open suctioning, administration of nebulized treatment, tracheostomy, and cardiopulmonary resuscitation\(^2\). Wearing personal protective equipment (PPE) is advised in order to prevent healthcare workers (HCW) from contracting COVID-19. An important part of PPE is facemasks and respirators. Surgical facemasks and Filtering Facepiece Particle (FFP) respirators are used by HCW, depending on the risk of producing aerosols while performing medical procedures. In the Netherlands surgical facemask type IIR and FFP1 respirators are used at COVID-wards. Use of FFP2 respirators is reserved for medical procedures with a high risk of aerosol formation. On 11 March 2020, the World Health Organization declared SARS-CoV-2 a pandemic. The SARS-CoV-2 pandemic has led to world-wide scarcity of PPE, including surgical facemasks, FFP1 respirators.
and FFP2 respirators. The Dutch National Institute for Public Health and the Environment (RIVM) has approved reuse of FFP2 respirators after hydrogen peroxide and steam sterilization treatment. Surgical masks and FFP1 respirators were not tested. Not every healthcare institution has access to these disinfection facilities. Droplets are mostly filtered at the exterior of the FFP respirators, but aerosols with infectious virus particles are also trapped in the inner layers of the respirators. Because we propose non-personalized reuse of respirators, we think it is important to show that we can thoroughly disinfect the inner layers of FFP respirators by UVC as well. In this paper we substantiate ultraviolet C (UVC) decontamination and reuse of surgical IIR facemasks, FFP1- and FFP2 respirators as a scalable solution to mitigate shortage.

Microbiology
Coronaviruses (CoVs) belong to the order of Nidovirales, the family of Coronaviridae and the subfamily of Coronavirinae. The Coronavirinae subfamily consists of: alphacoronavirus, betacoronavirus, gammacoronavirus and deltacoronavirus. SARS-CoV-2 belongs to the betacoronavirus subfamily. SARS-CoV and MERS-CoV also belong to the betacoronavirus subfamily. CoVs are enveloped viruses with a ± 30 Kb large + single stranded RNA genome. The genome of SARS-CoV-2 has a 79,5-82 % homology to SARS-CoV. UVC susceptibility studies of SARS-CoV have been described in literature. Therefore, we can infer the UVC susceptibility of SARS-CoV-2. In two studies of respiratory materials of COVID-19 patients the viral load was determined. A total of 27 patients was analyzed. The maximal viral load of oro- and nasopharyngeal samples was 1,5 × 10^7 and 7,11 × 10^8 copies/ml respectively. Therefore, we think it is important to achieve an 8-log reduction at the exterior side of the facemask.

UVC interaction
UVC is also used to disinfect water and surfaces. The germicidal effect of UVC is the result of a photolytic effect eliminating DNA and RNA replication potential. The absorption spectrum of nucleotides, which make up DNA and RNA, has a characteristic peak at 260 nm, indicating a strong UVC interaction. RNA is known to be more sensitive to UVC than DNA because the Uracil nucleotide that in RNA replaces the DNA Thymine nucleotide has a stronger UVC absorbance. In Figure 1,
both absorption spectra are displayed. The vertical line corresponds to a wavelength of 253.7 nm which is the output of a low-pressure UVC source. Single-stranded RNA, e.g. coronaviruses, is more sensitive to UVC than double-stranded RNA and DNA.

![Comparison of Thymine and Uracil UVC absorption spectra](image)

**Figure 1 – Comparison of Thymine and Uracil UVC absorption spectra (Kowalski)**

**Coronaviruses and UVC disinfection**

The effectiveness of UVC disinfection on various microorganisms has been well documented in literature and is reported as a microorganism specific D90 dose value expressed in J/m². The D90 dose value is a measure for the UVC tolerance and specifies the dose level at which 90% of a specific type of microorganism is inactivated. The UVC interaction is considered a stochastic effect in which a subsequent D90 dose exposure will subsequently affect 90% of the remainder of the microorganism.

A reference D90 dose value for SARS-CoV-2 can be derived from published D90 dose values for different coronaviruses. An overview table of UVC susceptibility studies in Coronavirus is available in the literature, this table is included below. It is unclear why the last 2 studies (Kariwa 2004, Darnell 2004) show a deviating value,
for completeness all results are shown here. However, cyanobacteria (blue green algae) which have chlorophyll pigments that absorb UV light are known to have a D90 of 3000 J/m². Since CoVs do not contain such pigments a D90 value in this range is unlikely.

Table 1: Summary of ultraviolet studies on coronaviruses

| Microbe                        | D90 J/m² | UVGI k m²/J | Media | RH % | Dia. μm | Base Pairs kb | Source    |
|-------------------------------|----------|-------------|-------|------|---------|---------------|-----------|
| Coronavirus                   | 3        | 0.37700     | Air   | 50   | 0.113   | 30.738        | Walker 2007 |
| Coronavirus                   | 7        | 0.32100     | W     | Wat  | 0.113   | 30.738        | Weiss 1966  |
| Coronavirus (SARS)            | 9        | 0.25340     | W     | Wat  | 0.113   | 29.751        | Duan 2003   |
| Coronavirus (SARS)            | 226      | 0.01000     | W     | Wat  | 0.113   | 29.751        | Kariva 2004 |
| Coronavirus (SARS)            | 3046     | 0.00076     | W     | Wat  | 0.113   | 29.751        | Darnell 2004|
| Genomic Prediction            | 7        | 0.3289      | W     | Wat  | 0.113   | 29.751        | Kowalski 2015|

Based on a mathematical genomic prediction model for the SARS-CoV genome the expected D90 for SARS-CoV is calculated at 7 J/m². This is consistent with the first 3 studies. These experimental assays and the genome-based prediction show that in general coronaviruses are sensitive to UVC. For comparison, some reference values for other microorganisms are given in table 2. A conservative uttermost estimate for a D90 for SARS-CoV-2 is taken to be 30 J/m².

Table 2: Reference D90 values

| Microbiological group | Type                      | D90 in J/m² for 254 nm |
|-----------------------|---------------------------|------------------------|
| Bacteria              | *Staphylococcus aureus*   | 26                     |
| Bacteria              | *Mycobacterium tuberculosis* | 60                   |
| Bacteria              | *Pseudomonas aeruginosa*  | 55                     |
| Bacteria              | *Clostridium tetani*      | 120                    |
| ssRNA virus           | Influenza                 | 36                     |
| ssRNA virus           | MS2-coliophage            | 186                    |

Influence of UVC on the effectiveness of the FFP respirators

The effects of UVC on similar FFP respirators have been published in 2015\textsuperscript{16}. This study found that high levels of UVC exposure to FFP respirators led to a small increase in particle penetration (up to 1.25%) and had little effect on the flow resistance. A more pronounced effect was seen on the bursting strengths of the respirator materials. In this study the particle penetration and airflow resistance have been examined at dose levels over 400 times higher and material strength over 1000
times higher than the dose clinically achievable. No significant changes to the respirator effectiveness are expected at the dose levels applied clinically for UVC disinfection.

**Methods**

**Transmission measurement by UVC indicator**
A robotic UVC setup designed for disinfection of surfaces is used as UVC source. The setup consists of 24 UV-C lamps of the type TÜV PL-95W / 4P HO 1CT (95W High Output Rated / 27W UVC Emission). Each lamp has an output of 2.5 W / m² UVC at a distance of 1 meter. A first transmission experiment is performed based on a UVC dose indicator strip from Intelligo Technologies by covering the indicator strip with a M3 FFP1 Aura™ 1861+ respirator and applying a 30-minute exposure to the UVC source at approximately 1 meter from the closest lamp.

**UVC transmission measurements by UVC meter**
Further transmission measurements have been performed with a calibrated UV meter and data logger (model: UV-Touch) All measurements were taken at 1 meter from the same robotic UVC setup. Measurement values are corrected for a background intensity and all intensity measurements are averaged over 1 minute. One item of several models of masks and respirators have been tested i.e.: a white surgical facemask (medline), a KN95 respirator, air PROtm(Kolmi), surgical facemask blue (3M), FFP1 Aura™ 1861+ (3M) and a FFP2 Aura ™ 1862+ (3M).

**S. aureus serial dilution UVC disinfection experiment**
We made factor 10 serial dilutions of *S. aureus* ATCC 25923 suspension in 0.9% NaCl. We prepared 6 Surgical IIR facemasks, 6 FFP1 respirators (3M Aura 1861+), 6 FFP1 respirators (Kolmi purple) and 4 FFP2 respirators (3M Aura 1862+). We made an incision from the face-side of the FFP1- and FFP2 respirators and placed 1.5 x 2 cm sterile wound gauze (Cutisorb) after the first two polypropylene layers in the middle of the FFP respirator. 1.5 x 2 cm of sterile wound gauze was fixated at the exterior-side of the surgical mask. We inoculated 10 µl of *S. aureus* dilution in several concentrations within 1 cm² of the gauze (polypropylene is hydrophobic material). We closed the flap of the FFP respirator. Thereafter we irradiated the facemasks with 6 UVC lamps at 1-meter (type TÜV PL-L 95W/4P HO 1CT) 95W High Output Rated 27W UVC emission. Each lamp has an output of 2.5 W/m² UVC at 1-meter. We irradiated each side of the masks for 20 minutes. After 40 minutes of irradiation we collected the wound gauze and incubated the gauzes in thioglycollate broths (Oxoid) at 36°C O₂ for a total of 40 hours. After 16 hours and 40 hours we visually inspected the broths
and subcultured 10 µl of thioglycollate broth on sheep blood agars (Oxoid) at 36°C O2 overnight. The next day we inspected the blood agars for growth and determined the bacteria by MALDI-TOF MS (Bruker).

**Results**

**Transmission measurement by UVC indicator**

A dose of approximately 10 mJ / cm² behind an M3 FFP1 mask is observed at an estimated applied dose to the mask of 2700 mJ / cm² based on a 30-minute irradiation with 6 lamps, each with an output of 2.5 W / m² at the position of the mask. A first approximation of the transmission of the UVC transmission of the mask therefore is in the order of 0.37%.

![UVC Dose Indicator](image_url)

**Figure 2 - Left the dose measured behind the mask. On the right a non-irradiated indicator. The MRSA color correlates to 10 mJ/cm²**

Figure 2 shows that the dose distribution on the irradiated indicator is not homogeneous. Due to the material properties of this mask, more shielding takes place in some places in the mask than in others. In addition, a considerable scatter fraction can also be expected in practice.

By neglecting a scatter fraction and assuming an exponential decay of the dose throughout a homogenous material a minimal dose level in the middle of the material can be estimated. The dose at 50% material depth is the square root of the transmission at 100% of the material. Exposing the material equally from the front and back side will furthermore double the dose in the middle of the material. An estimated transmission fraction of 0.37% will yield a dose in the middle of the material of 2*0.0037¹/² = 0.122 or 12.2% from the entrance dose as is visualized in figure 3.
Figure 3. Visualization of UVC dose in homogenous material without scatter effects.

UVC transmission measurements by UVC meter

An UVC intensity of 18.1 W/m² was measured as an entrance dose for the FFP respirator and the background intensity was measured to be 0.002 W/m². The measured UVC intensities that pass through the FFP respirators and masks have been corrected for the background intensity and are listed in table 3. The transmission is expressed as the percentage of the exiting UVC intensity compared to the entrance intensity. A dose in the middle of the material is calculated assuming an exponential decay and no scatter effects. The UVC transmission for the FFP2 Aura ™ 1862+ could not be measured by the UVC meter and will be below 0.001 W/m² corresponding to an intensity in the middle of the mask lower than 1.5 % from the entrance intensity.
Table 3: UVC transmission for different FFP respirators

| Respirator type                        | Measured intensity W/m² | % transmission | Calculated intensity in the middle of the mask |
|----------------------------------------|-------------------------|----------------|-----------------------------------------------|
| medline / white surgical facemask      | 1.365                   | 7.55%          | 55.0%                                         |
| KN95                                   | 0.931                   | 5.15%          | 45.4%                                         |
| Kolmi air PRO™                          | 0.308                   | 1.71%          | 26.1%                                         |
| Surgical facemask blue 3M              | 0.126                   | 0.70%          | 16.7%                                         |
| FFP1 Aura™ 1861+, 3M                   | 0.008                   | 0.05%          | 4.3%                                          |
| FFP2 Aura™ 1862+, 3M                   | <0.001                  | -              | -                                             |

S. aureus serial dilution UVC disinfection experiment

In table 4 we see that the process controls, number 1,7,11 and 17 are positive. These were treated the same except for UVC irradiation. The negative controls remained negative except for number 16. Number 16 showed Bacillus subtilis (a known contaminant). FFP2 respirators show no growth after an inoculation of 2.10^7 (or lower) S. aureus CFU/ml. We obtain at least a 7-log reduction of S. aureus in the middle of the FFP2 respirator. FFP1 respirators show no growth after an inoculation of 2.10^8 (or lower) S. aureus CFU/ml. We obtain at least an 8-log reduction of S. aureus in the middle of the FFP1 respirators. The surgical IIR masks show growth of S. aureus an inoculation of 2.10^9 CFU/ml, but does not show growth after an inoculation of 2.10^8 (or lower) S. aureus CFU/ml. We obtain an 8-log reduction of S. aureus at the exterior of the surgical IIR masks.
Table 4. UVC irradiation of facemasks inoculated with S. aureus.

| Number | Mask type     | CFU/ml | Growth on blood agar – 40 hours |
|--------|--------------|--------|---------------------------------|
| 1      | No irradiation FFP1 3M | 2.10^8 | Growth S. aureus                |
| 2      | FFP1 3M      | 2.10^8 | No growth                       |
| 3      | FFP1 3M      | 2.10^7 | No growth                       |
| 4      | FFP1 3M      | 2.10^6 | No growth                       |
| 5      | FFP1 3M      | 2.10^5 | No growth                       |
| 6      | FFP1 3M      | 0.9% NaCl | No growth                      |
| 7      | No irradiation FFP2 3M | 2.10^7 | Growth S. aureus                |
| 8      | FFP2 3M      | 2.10^7 | No growth                       |
| 9      | FFP2 3M      | 2.10^6 | No growth                       |
| 10     | FFP2 3M      | 0.9% NaCl | No growth                      |
| 11     | No irradiation Surgical IIR | 2.10^9 | Growth S. aureus                |
| 12     | Surgical IIR | 2.10^9 | Growth S. aureus                |
| 13     | Surgical IIR | 2.10^8 | No growth                       |
| 14     | Surgical IIR | 2.10^7 | No growth                       |
| 15     | Surgical IIR | 2.10^6 | No growth                       |
| 16     | Surgical IIR | 0.9% NaCl | Contamination: Bacillus subtilis |
| 17     | No irradiation FFP1 Kolmi | 2.10^8 | Growth S. aureus                |
| 18     | FFP1 Kolmi   | 2.10^8 | No growth                       |
| 19     | FFP1 Kolmi   | 2.10^7 | No growth                       |
| 20     | FFP1 Kolmi   | 2.10^6 | No growth                       |
| 21     | FFP1 Kolmi   | 2.10^5 | No growth                       |
| 22     | FFP1 Kolmi   | 0.9% NaCl | No growth                      |

Discussion

A conservative estimate of the exposure necessary to thoroughly disinfect the FFP respirators can be calculated based on the measured transmission and assumed D90 value. For example, the FFP1 M3 respirator used in these experiments has a 4.3% dose in the middle of the FFP1 respirator. In order to yield an 8-log reduction of SARS-CoV-2 with the estimated D90 of 30 J/m² a dose of 8x30 or 240 J/m² in the middle of the FFP1 respirator would be necessary. The externally applied dose would have to be greater than 240 / 0.043 = 5580 J/m². It is possible to achieve such a dose level clinically in our setup in approximately 5 minutes of exposure per side. In practice we use a multiple of this exposure time to account for possible effects such as microshielding and scatter.

We showed an 8-log reduction of S.aureus in the middle of FFP1 respirators (3M and Kolmi) and the exterior of surgical IIR facemasks. Furthermore, we showed a 7-
log reduction of \textit{S. aureus} in the middle of FFP2 respirators. The reduction of SARS-CoV-2 would be similar or better depending on the true D90 value which is expected to be lower than that of \textit{S. aureus}.

Despite the limit of detection of transmission of UVC through FFP2 respirators it is apparently sufficient to irradiate the FFP2 respirators from both sides to attain an adequate UVC dose within the FFP2 respirator. The limit of detection lies at 1.5% of the entrance dose in the middle of the FFP2 respirator. In theory it would still be possible to achieve an 8-10-log reduction for \textit{S. aureus} in the FFP2 respirator. We did not test higher concentrations than $2 \times 10^7$ CFU/ml, because our transmission experiments initially led us to believe FFP2 respirators were not suitable for UVC disinfection. The surgical IIR facemask shows growth at an inoculation of $2 \times 10^9$ \textit{S. aureus} CFU/ml after UVC irradiation of 20 minutes at either side of the mask. This concentration of bacteria normally only occurs in the intestines of patients. There could be other factors involved in that limits UVC disinfection for extremely high concentrations. We do advice to exclude all visibly soiled facemasks for UVC disinfection. Literature shows that no impairment of the effectiveness is expected for different respirators at the applied UVC dose levels for disinfection\textsuperscript{16}. In our experience individual fittest do not lead to rejection of the disinfected masks and respirators. The UVC disinfection procedure can be repeated multiple times. We repeat this procedure up to three times.

\textbf{Conclusion}

We have shown that it is possible to obtain sufficient UVC dose throughout surgical facemasks, type IIR, FFP1 and FFP2 respirators to achieve respectively an 8-log and 7-log \textit{S. aureus} reduction. The UVC sensitivity of \textit{S. aureus} is similar to that of SARS-CoV. Due to a high homology of SARS-CoV-2 to SARS-CoV we expect similar disinfection results. Our used UVC dose is not expected to affect respirator material and efficacy. Therefore face-fit will not change with repeated UVC treatments. The process of reusing facemask and respirators should be carefully monitored. Masks and respirators should be collected safely, visually inspected and marked for the number of times they are UVC treated.

The masks and respirator stability under UVC exposure and the widespread application and availability of UVC lamps provide a scalable method for disinfection.
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