COMPATIBILITY AND EFFICACY OF VAPORIZED HYDROGEN PEROXIDE TECHNOLOGY TO DECONTAMINATE REUSABLE PERSONAL PROTECTIVE EQUIPMENT

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Abstract: This study aimed to evaluate VHP as a decontamination method for decontaminating reusable PAPR; determine if PAPR is compatible with VHP; and determine how long it takes for hydrogen peroxide to off-gas for the mask to be safe for reuse. Off-gassing validation study which consisted of doing three VHP decontamination processes followed by three Off-Gassing Tests to confirm the efficacy of the VHP cycle and time required for the VHP concentration to reach 0 ppm. HALT, which consisted of exposing the PAPR to 50 VHP decontamination cycles followed by a Visual Appearance Test to determine if the VHP process impacted the PAPR material’s visual properties. The results from this study show that VHP is an effective and repeatable process for decontaminating PAPR, and the Off-Gassing

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PUBLIC INTEREST STATEMENT

Powered Air-Purifying Respirators (PAPR) are reusable Personal Protective Equipment (PPE) designed to provide respiratory protection. PAPR’s intricate design and diverse materials make them difficult to decontaminate between uses, posing an infection risk. Vaporised Hydrogen Peroxide (VHP) has been widely used to decontaminate disposable PPE during COVID-19; however, there is limited evidence on the compatibility of VHP with reusable PPE. This study focused on evaluating the efficacy, safety, and compatibility of using VHP for decontaminating PAPR. This was achieved by performing: 12 VHP Off-Gassing Tests, and a Visual Appearance Test (including Colour, Opacity, and Photographic Appearance Tests) before and after 50 VHP cycles. Results show VHP as an effective technology for decontaminating PAPR, and that exposing a PAPR to 50 Decontamination Cycles does not change the PAPR appearance in a significant way.
Test shows that with the appropriate off-gassing set up, it is possible to off-gas one PAPR within 8 hours of completing the decontamination cycle. The results from the Visual Appearance Test show that exposing a PAPR to 50 decontamination cycles does not present significant changes in the PAPR material’s colour, opacity, or overall visual appearance.

Subjects: Cleaning & Sterilization; Nondestructive Testing; Industrial Textiles; Infectious Diseases

Keywords: Powered Air-Purifying Respirators (PAPR); Personal Protective Equipment (PPE); decontamination; Vaporised Hydrogen Peroxide (VHP); infection control; compatibility

1. Introduction

1.1. Background
COVID-19, a disease caused by the SARS-CoV-2 virus, was declared a pandemic in March 2020 (WHO, 2020). At an early stage, it was identified that the primary route of transmission of the virus is through inhaling aerosols and/or droplets exhaled by an infected person if close contact occurs (Thaper et al., 2021) and, consequently, the use of PPE became one of the main measurements to control the spread of SARS-CoV-2. There is a wide variety of options for respiratory protection (Licina & Silvers, 2021), with disposable and reusable formats to choose from and, while disposable respirators such as N95 face masks have gained popularity during the pandemic, reusable alternatives like Powered Air-Purifying Respirators (PAPR) have recently started to be considered as a more environmentally friendly option.1,2,3

As a response to the widespread use of disposable PPE and supply chain disruptions across the globe, numerous studies emerged to decontaminate disposable items, using technologies such as vaporised hydrogen peroxide, hydrogen peroxide gas plasma, ultraviolet germicidal irradiation, moist heat, ethylene oxide, gamma irradiation, microwave-generated steam, and 70% or higher ethanol solution (Cramer et al., 2021; Thaper et al., 2021). While most decontamination approaches appeared as a temporary solution for the shortages on disposable items, some of those technologies could have great value if implemented for reusable items during both pandemic and non-pandemic scenarios. In particular, Vaporised Hydrogen Peroxide (VHP) has shown to be successful in decontaminating various types of masks and respirators (Schwartz et al., 2020; Saini et al., 2020).

There is evidence in the literature regarding the efficacy of decontamination technologies against pathogenic microorganisms; however, there is limited evidence of the compatibility of such technologies with the different materials used to manufacture PPE. In addition, some decontamination methods can leave chemical residues that need to be removed before being able to reuse the protective item, a topic that has not been appropriately addressed in recent studies. To permanently implement a new decontamination technology in healthcare institutions, proper evidence needs to be generated to ensure that such processes will achieve efficacy against harmful microorganisms, will be compatible with PPE materials, and more importantly that the decontaminated items will be safe to use.

The purpose of this study was to 1. Demonstrate VHP as an effective, repeatable decontamination technology suitable for PAPR; 2. Determine the required time for off-gassing PAPR after VHP decontamination process to eliminate residual chemical hazards; and 3. Determine if the exposure of PAPR to repeated decontamination cycles influenced PAPR material’s visual properties and overall post-decontamination integrity.
1.2. Respiratory protection

The main purpose of using a respirator is to prevent the inhalation of harmful airborne substances. To meet such purpose, respirators usually include an enclosure to cover the nose and mouth or the entire face or head (Szeinuk et al., 2000). Respirators are classified by the method they use to deliver their protective action: air-purifying respirators filter airborne particles, and atmosphere-supplying respirators supply clean air to the wearer (Park, 2020). Air-purifying respirators can be further classified into filtering facepiece respirators (FFRs), elastomeric facepiece respirators, and PAPR. Among these three categories, FFRs are the most used because of their disposable nature. FFRs are categorised according to their filtering efficiency, and their main limitation is that, in order to provide proper protection, they must tightly fit the face of the user (Park, 2020).

Prevalence towards the use of disposable respiratory protection creates a considerable amount of waste that has been significantly increased since the onset of the pandemic. While there is a current trend towards adopting reusable PPE as a more sustainable option, there are still concerns from an infection control perspective. Reusable items can become difficult to disinfect because of the porous nature of their materials and, while there are effective formulas to remove the pathogenic microorganisms, they can pose a negative effect on the integrity of the item. Unfortunately, there is limited evidence in the literature regarding compatibility studies between disinfection technologies and reusable PPE.

As a response to the PPE shortages experienced from the onset of the pandemic, several research studies emerged exploring decontamination methods for decontaminating disposable PPE. For decontaminating FFRs in particular, the National Institute for Occupational Safety and Health (NIOSH) performed a literature review on the potential decontamination methods for FFRs that would result effective in reducing the pathogenic burden, without harming the fit or filtration performance, and not leaving residual chemical hazards, and the outcome was the following were identified as the most promising potential methods for decontaminating FFRs: ultraviolet germicidal irradiation, vaporous hydrogen peroxide, and moist heat (Centers for Disease Control and Prevention, 2020).

However, such methods required adaptation since most decontamination technologies were originally created for healthcare environments and hard surfaces. While currently there are several decontamination methods that show efficacy against the pathogenic microorganisms present in the healthcare environment, not all of them are suitable for using on PPE items. For example, liquid disinfectant products used through immersion techniques, or wipe-down formulations are not only time-consuming but can also negatively affect the PPE materials’ integrity (Saini, Sikri, Dhingra Batra, Kalra, & Gautam, 2020).

Any decontamination technology that is being considered as a suitable option for the disinfection and reuse of PPE during future pandemic situations should be compatible with different types of PPE items to suit diverse user needs, as well as have solid quality assurance controls (Saini et al., 2020). A study by Thaper, Fagen & Oh reviewed the advantages and disadvantages of the following decontamination methods for their use on respiratory protection items: Ethanol; Ultraviolet germicidal irradiation (UVGI); Microwave-generated steam (MGS); Vaporized Hydrogen Peroxide (VHP); and Hydrogen peroxide gas plasma (HPGP). The analysis considered decontamination efficacy, respirator function, and feasibility. The authors reached the conclusion that Vaporised Hydrogen Peroxide (VHP) is the most promising method based on the high biocidal efficacy on FFRs and is also currently the most promising option for PAPRs since it shows the same efficacy as other technologies, but results in considerably less damage to the materials and components of the equipment (Thaper et al., 2021).
1.3. Decontamination using vaporised hydrogen peroxide (VHP)

Hydrogen peroxide (HP) is an oxidising agent, and its vapourised form, Vaporised Hydrogen Peroxide (VHP), is used as a biocide as it is capable of breaking DNA/RNA strands via oxidation (Saini et al., 2020).

Over the last few decades, commercially available VHP technologies have been successfully implemented in the healthcare industry, in applications ranging from product decontamination for laboratory and medical equipment, to room decontamination for hospital wards and pharmaceutical manufacturing buildings (Linley et al., 2012). For such applications, VHP has shown great efficacy against a wide range of microorganisms, including (but not limited to) endospores, Gram-positive and Gram-negative cells, DNA and RNA viruses, and fungi (Linley et al., 2012).

During the onset of COVID-19 it was evident that the use of VHP to decontaminate PPE needed to be evaluated. Several studies addressed the use of VHP to decontaminate respiratory devices with a strong focus on N95 respirators (Cramer et al., 2021; Al-Hadyan et al., 2021), and other studies had an even broader approach including gown decontamination (Saini et al., 2020). A study done on N95 respirators showed that after 50 cycles the item retained its functional and fit properties (Schwartz et al., 2020). Another study showed that FFP2 face masks exposed to two hydrogen peroxide processes were able to remain their shape as well as retain particles; however, those masks exposed to three and four cycles showed deformation, which compromised their usability (Dutch National Institute for Public Health and the Environment (RIVM), 2020). While these studies provide important insights about the impact of VHP on PPE, they focus on disposable items typically made of nonwoven materials. There is a need for further studies on reusable materials in order to properly analyse the impact of VHP on woven and knitted textiles.

An important aspect of using VHP that needs to be considered is the potential chemical residues that can remain on the surface of the treated item. A study analysing hydrogen peroxide residue levels identified high levels of hydrogen peroxide in the gas phase and on the surface of N95 masks, highlighting the potential risks of respiratory and skin contact (Kumkrong et al., 2021). The study also identified that the hydrogen peroxide concentration found on the mask surface was higher than the concentration measured within the confined space.

1.4. Powered-air purifying respirators (PAPR)

A Powered-Air Purifying Respirator (PAPR) is a type of respiratory protection that consists of using a pump to force air through filter cartridges or canisters and into the breathing zone of the wearer. As a result, air flow is created inside either a tight-fitting facepiece or loose-fitting hood or helmet (CDC, 2020).

Due to their high level of respiratory protection, PAPRs are usually the preferred option for healthcare workers who need to perform high-risk aerosol generating procedures. Also, their design provides better comfort for the wearer, making them desirable for performing complex airway procedures, and also for healthcare workers who fail a fit test for disposable respirators (Chakladar et al., 2021).

PAPRs offer several benefits: they provide a high level of protection, they have less of an impact on the user’s breathing when compared to other protection devices, they do not require fit testing, they are reusable, and they have a loose-fit headgear that makes them suitable for users with a limited amount of facial hair (Park, 2020; Licina & Silvers, 2021). However, the use of PAPR has some disadvantages: due to their reusable characteristics, PAPRs require a care regime of cleaning, decontamination, storage, and battery maintenance that can prove challenging from a logistical point of view (Licina & Silvers, 2021).
PAPRs require proper cleaning and disinfection between uses, a task that must be performed by trained individuals, and also there are special doffing processes that need to be followed in order to avoid the risk of contamination (Park, 2020). UK infection control guidelines recommend that PAPRs should be cleaned and disinfected with disinfection wipes but, unfortunately, such guidelines do not address how hoods and air supply hoses should be processed (Chakladar et al., 2021).

Some research studies have addressed the infection control challenges associated with the use of PAPRs. A study conducted by Chakladar et al. performed a swab test on 25 PAPR previously used but cleaned and disinfected. As a result, bacteria and fungi were found on the interior components of the PAPRs’ hoods and air supply hoses. Researchers concluded that PAPRs have the potential to act as fomites, with the risk of cross-infection of wearers and patients, and that current PAPR disinfection guidelines might not be effective for use in high-risk healthcare environments (Chakladar et al., 2021).

2. Materials and methods

2.1. Study overview

In summary, the following tests were performed as part of the study:

- **Decontamination process:** decontamination of the PAPR was performed inside a sealed, cladded 12.1 m³ ProXpod chamber, using Vaporised Hydrogen Peroxide (VHP). Cycle parameters after each decontamination process were recorded. Standard Biological and Chemical Indicators were used during the test.

- **Post VHP Off-Gassing Test:** to determine the time taken for the VHP concentration to reach undetectable levels (i.e., 0 ppm) after decontamination, Off-Gassing Tests were performed. This consisted of monitoring the PAPR periodically using a gas monitoring unit to detect H₂O₂ levels.
  - As part of the Off-Gassing Test, preliminary tests (15 cycles) were performed to determine the best off-gassing scenario that would lead to a lower Off-Gassing time.
Highly Accelerated Life Cycle Test (HALT): to simulate the repeated decontamination, 50 continuous decontamination cycles were performed.

Visual Appearance Test: Visual Appearance Test was performed before and after 50 cycles of the HALT to determine if the test caused any physical changes. Details about possible visual spots, marks, colour change, or material modification were recorded. In addition, colour and opacity measurements were taken.

2.2. Materials and equipment

2.2.1. Test item
The test items used for this study consisted of two commercially available CleanAir CA-1+ Powered Air-Purifying Respirators (PAPR) (see Figure 1), with the following components: a visor made of cellulose propionate, a textile part made of laminated polyethylene (PP+PE), and a head harness made of polypropylene. The limited amount of test items is a result of material constraints and limited resources during the pandemic.

The two PAPR were labelled as follows: PAPR 01 and PAPR 02. To ensure that no additional variables were introduced during the test process that could have affected the Visual Appearance Test results, PAPR 01 was used to perform the Decontamination Process and Off-Gassing Test; and PAPR 02 was used to perform the HALT Test and Visual Appearance Test.

2.2.2. Vaporised hydrogen peroxide (VHP) decontamination
A ProXcide™ decontamination system was used to perform the Vaporised Hydrogen Peroxide (VHP) decontamination. The system is fully automated and uses ultrasonic vaporisation technology to create microscopic droplets of hydrogen peroxide that are forced-evaporated to create hydrogen peroxide vapour. The ProXcide™ decontamination system holds a hydrogen peroxide bottle with a 7.5% Hydrogen Peroxide solution that fits into the main unit.

Decontamination cycles were carried out using Inivos’ ProXpod decontamination chamber with the following dimensions: 2,400 mm × 2,400 mm × 2,100 mm (W×D×H) and door size 838 mm × 1,981 mm. The ProXpod was fitted with four wire shelving units to help position the test items. To help define the Biological and Chemical Indicator locations, each unit was assigned a number and each individual shelf was also given a number, as seen in Figure 2.
2.2.3. Biological and chemical indicators

Mesa Labs G. stearothermophilus HMV-091 1.8 × 10⁶ CFU per stainless steel carrier Biological Indicators (BI) were used to confirm the microbiological efficacy of the decontamination test cycle across different parts of the room, ensuring that worst-case positions were assessed. Previous studies showed that the use of bacterial spores provides a good estimation of the efficacy of decontamination processes since spores can be more resistant than viruses (Cramer et al., 2021). The microorganism selection for this study was also based on previous evidence suggesting that Geobacillus stearothermophilus can be difficult to kill when using VHP (Cramer et al., 2021).

For this study, a total of 12 BIs were used on each decontamination cycle, placed on chamber locations that provided a challenge to the decontamination technology. For the VHP decontamination technology, it is more likely to achieve efficacy the closer the indicator is placed to the main decontamination unit. In this study, considering that the unit was placed in the middle of the test chamber, the most challenging locations were the upper and lower corners. In addition, positive controls (unexposed disc) were used to show that the growth conditions were suitable and none of the reagents or BI discs were faulty; and negative controls (blank tube of media) were used to highlight any possible cross contamination picked up during the process.

Excelsior Chemical Indicators (CI) were used to demonstrate gas penetration at identified worst-case positions (same as those used for the BI), by showing a colour transition from brown to green after successful exposure to vapourised H₂O₂. In total, 12 BI and 12 CI were used in the following locations (see, Figure 2): 1 T, 1 M, 1B, 2 T, 2 M, 2B, 3 T, 3 M, 3B, 4 T, 4 M, 4B. In addition, positive and negative controls were used as follows: 1 positive control stainless steel carrier G. stearothermophilus spore token, and 1 negative control broth (uninoculated broth) outside the chamber.

2.3. Decontamination procedure and BI/CI processing

For each Decontamination Cycle, the ProXcide™ was placed in the centre of the ProXpod chamber and the PAPR 01 was placed on the middle shelf between shelving units 1 and 3. Biological Indicators were placed in labelled sterile petri dishes and positioned inside the ProXpod with the lids open at the specified locations. Chemical Indicators were also placed at the specified locations using labelled tape. After placing all the test items, the ProXcide unit was operated as per the user manual to run a complete decontamination cycle. In total, 3 decontamination cycles were performed, each of them having a subsequent Off-Gassing Test.

After the decontamination cycles, BIs were placed inside a sterile petri dish with the lid closed and taken to a class II microbiological safety cabinet. Inside the cabinet, using disposable sterile forceps, an unexposed BI was placed directly into a Mesa Labs Releasat media tube, acting as positive control. Each of the BI discs that were retrieved from the chamber (exposed to VHP) were placed into the corresponding Releasat media tube, and one Releasat tube without any BI disc was used as Negative Control. After lightly loosening the broth lids, media tubes were incubated under aerobic conditions at 60°C and checked for colour changes at both 24 hours and 7 days, recording the results at each time. A change in the original colour of the media indicates the presence of microorganism growth. Mesa Labs Releasat media changes from purple to yellow if growth is detected.

2.4. Off-Gassing test

One of the critical parts of using VHP is dedicating the appropriate amount of time for the decontaminated items to naturally release the VHP traces that might have remained on the different item materials after the cycle. This process can be defined as Off-Gassing, and it can have varied lengths depending on variables such as item size and materials, chamber size, and hydrogen peroxide concentration. This study focused on estimating the time required for a PAPR to Off-Gas after a ProXcide decontamination cycle. For decontaminated items to be safe to use after VHP
decontamination, the VHP concentration must reach 0 ppm which represents undetectable levels of hydrogen peroxide. This was measured by performing an Off-Gassing Test that consisted of periodically monitoring the test item and its components using a gas monitor. Special interest was placed on determining the total time required for the PAPR to reach to 0 ppm after each decontamination process.

Preliminary studies were performed to determine the best off-gassing set up that would result in the shortest off-gassing time. This was determined by adjusting environmental parameters and trying different configurations, including the following scenarios: well-ventilated area, use of fan, use of portable heater, use of fan and portable heater. During the first part of the preliminary study phase, off-gassing measurements were taken at hourly intervals to have a better understanding of the off-gassing times on the different PAPR materials. Early on the preliminary studies, it was identified that performing the off-gas in a well-ventilated area without modifying environmental conditions would mean having more than two days of off-gassing. External variables were then introduced: heat and air circulation, and using a closed chamber for the off-gassing process to maintain the environmental conditions. This last set up considerably improved the results obtained, therefore, was selected as the preferred set up.

The off-gassing station selected for the off-gassing process consisted of a ProXpod chamber, in which the PAPR was placed facing upwards to increase the aeration on the internal components, using a portable heater set at 28°C and a fan at maximum speed on tilting mode, and closing one of the ProXpod doors while leaving the other door slightly open.

The Off-Gassing Test was conducted within 30 minutes after the completion of each of the 3 decontamination cycles above mentioned. Measurements were taken in a well-ventilated area, and at the following locations of the PAPR: Exterior Textile; Interior Textile; Head Harness Textile; Interior Elastic; Exterior Plastic Visor. For each location, three measurements were taken from different areas using different gas meters. Off-gassing measurements were done using a Porta Sens D16 III H₂O₂ gas monitor with a H₂O₂ cell calibrated for 0–10 ppm. After taking all the required measurements, the PAPR was placed on the Off-Gassing station for 8 hours for the off-gassing process to take place. After the set time, the PAPR was retrieved, and another set of measurements was taken at the same locations.

For the off-gassing process to be considered finalised, all PAPR locations had to be labelled as completed. A complete label was assigned to each location only if, out of the three measurements performed, two showed 0.0 readings, with the additional requirement of not having any ppm measurement above 0.5.

2.5. Compatibility testing
Decontamination technologies must not only be effective and safe but must also preserve the treated item properties and integrity so that the PPE can be used for the intended purpose throughout its lifecycle. The compatibility test aimed to determine if exposing the PAPR to repeated decontamination cycles could have an impact on the item’s visual properties. To achieve such goal, Qualitative and Quantitative Visual Tests were performed before and after exposure to 50 decontamination cycles.

2.5.1. HALT
Highly Accelerated Life Cycle Test (HALT) simulates the repeated use of a product over the course of its entire lifetime. In this study, the HALT Test consisted of exposing PAPR 02 to a total of 50 VHP cycles. During the HALT Test the PAPR was placed on the middle shelf inside a ProXpod
Decontamination Chamber, the ProXcide™ unit was placed in the centre of the chamber, and a dehumidifier was placed near the door so that its screen could be monitored from the outside.

2.5.2. Visual test

Decontamination technologies can damage medical devices and healthcare surfaces, and changes in appearance are usually the first ones to be noticeable. To our knowledge, there is a lack of standards to assess the compatibility of PPE to decontamination technologies. However, there are published standards on related fields that are pertinent to be used as inspiration for selecting the right types of testing to perform that would allow to assess compatibility of items used for healthcare applications. In particular, standard ISO 21530:2004 titled “Dentistry—Materials used for dental equipment surfaces—Determination of resistance to chemical disinfectants” proposes a Visual Test as a way for determining the resistance to chemical disinfectants of dental equipment (International Organization for Standardization (ISO), 2018). Components of such Visual Test were used as reference for developing a custom Visual Testing plan specifically targeted to PAPR. However, since the methods on ISO 21530:2004 rely mostly on qualitative methods that could lead to issues related to the subjectivity of the visual examination, it was decided to also incorporate qualitative methods in this study to have a less subjective approach.

In summary, two types of Visual Tests were performed to determine if the HALT Test caused the appearance of the PAPR to change: a Quantitative Test, consisting of an Opacity Test and a Colour Test, and a Qualitative Test, consisting of a Photographic Appearance Test and Subjective Appreciation. Each of those tests were performed before and after exposure to the 50 HALT Cycles

2.5.2.1. Photographic appearance test. The test item (PAPR 02) was photographed under consistent lighting conditions in order to accurately record the visual appearance of the test item, and any changes in colour, opacity, spots, and/or marks were recorded. Pictures were taken at specified angles to capture the condition of the test item from different sides. Consistent lighting was achieved by using a Colour Assessment Cabinet (CAC) with a Daylight D65 Lamp as per the specification set in the BS 950–1:1967 “Artificial daylight for the assessment of colour”. The set up consisted of placing the test item inside the CAC and placing a Nikon D3500 DSRL Camera on a tripod in front of the CAC. In addition, a tilting table was used for taking pictures at different angles.

2.5.2.2. Subjective appreciation. A subjective analysis of the PAPR was performed to assess the overall condition of the test item and identify any possible faults and/or damage. This subjective analysis included performing a visual observation of the item, as well as handling and manipulating the item to assess the condition of the different materials. Any observations and comments were recorded.

2.5.2.3. Opacity test. Opacity readings were taken on the PAPR’s plastic visor using an NDT Tester—Opacity Meter PCE-RM 100. Measuring opacity was considered pivotal for a PPE item since any changes to the appearance of the visor may compromise its performance and therefore the safety of its user.

The opacity meter measures how much light is reflected through a material. The reflection of light is measured by placing an absolute white and absolute black working board behind a surface/material, and from this the degree of reflection is determined mathematically by the device. The outcome of the opacity measurement is a numerical value that can be used to assess the degree of opaqueness as follows: a 0 reading means the material is fully transparent, and a 100 reading means the material is fully opaque.

Opacity readings were taken in the following locations: Upper Left Corner; Upper Right Corner; Lower Left Corner; Lower Right Corner; and Centre.
2.5.2.4. **Colour test.** For the Colour Test, colour readings of different parts of the PAPR were taken using a PCE-XXM 20 Colorimeter. The measuring device is a tristimulus colorimeter, which operates by reflecting a light source at the surface of an object and then measuring the reflected light using a colour sensor. After the digitisation of the sensor data, the colour coordinates (L, a, and b) are calculated and displayed on the screen (see Figure 3).

Readings were taken in the following locations of the PAPR: Exterior Textile (Blue) Left at 3 locations; Exterior Textile (Blue) Right at 3 locations; Exterior Textile (Black) at 7 locations; Head Harness Textile at 3 locations; Interior Elastic at 6 locations.

Colour coordinates of the different PAPR sections were recorded before and after exposure to HALT cycles, and those values were then used to mathematically calculate the colour difference between the initial and final condition of the test item, as per BS EN ISO 11664–6:2016—Colorimetry. Part 6: CIEDE2000 Colour-difference formula:

\[
\Delta E_{00} = \left( \frac{\Delta L^'}{k_L S_L} \right)^2 + \left( \frac{\Delta C^'}{k_C S_C} \right)^2 + \left( \frac{\Delta H^'}{k_H S_H} \right)^2 + R_1 \left( \frac{\Delta C^'}{k_C S_C} \right)^2 \left( \frac{\Delta H^'}{k_H S_H} \right)^2
\]

After taking all the readings and calculating the colour difference values, results from each zone were averaged to obtain a single value for each of the zones.

3. **Results**

3.1. **Decontamination process using biological indicators and chemical indicators**

The three ProXcide decontamination cycles performed on this study prior to performing the Off-Gassing Test had a successful outcome, as seen on the Decontamination Process Reports created by the VHP unit (examples of such reports can be found in the Supplementary Material section).

The results from the BI, included in the Supplementary Material section, show that for the three decontamination cycles all BI passed the test. An example of the processing of BI and the media colour changes can be found in the Supplementary Material section.

Results from CI, included in the Supplementary Material section, show that all CI used on cycles 1, 2, and 3 passed the test. Photographs showing the colour change can also be found in the Supplementary Material section.

3.2. **Off-Gassing test**

The results from the Off-Gassing Test are shown in Table 1, and the complete raw data table can be found in the Supplementary Material section. Using an Off-Gassing set up consisting of a closed ProXpod chamber with a fan and portable heater, it was possible to off-gas PAPR #1 in 8 hours.

3.3. **Compatibility test**

3.3.1. **HALT**

As part of the Highly Accelerated Life Cycle Test (HALT), PAPR 02 was exposed to 50 ProXcide decontamination cycles. All 50 decontamination cycles performed were successful, as shown in the Decontamination Process Reports created after completion of each cycle.

3.3.2. **Qualitative tests**

3.3.2.1. **Photographic appearance.** The Photographic Appearance Test consisted of recording the visual appearance of test items under consistent lighting conditions using a DSLR camera. The
Figure 3. Photographic appearance results.

results for the Photographic Appearance Test performed before and after exposure to 50 HALT cycles are shown in Figure 4, which includes a subset of the different pictures taken.

3.3.2.2. Subjective appreciation. The second part of the Qualitative Test consisted of performing a Subjective Appreciation. A summary of the observations made before and after the 50 HALT cycles can be found in Table 2.
3.3.3. Quantitative test

3.3.3.1. Opacity test. As visor opacity is critical to the function of the PAPR, it is essential that the opacity is not affected by repeated decontamination. To evaluate this property, an Opacity Test was performed, taking measurements at different areas of the plastic visor surface. Results were automatically calculated by the Opacity Meter used. Results for the Opacity Test performed on the PAPR’s Plastic Visor before and after the HALT cycles are shown in Figure 5.

| Table 1. Off-gassing test results |
|-----------------------------------|
| PAPR location                     | Ppm after decontamination | Status after 8 hours |
|-----------------------------------|---------------------------|----------------------|
| Plastic visor                     | Cycle 01 | Cycle 02 | Cycle 03 | Cycle 01 | Cycle 02 | Cycle 03 |
| Plastic visor                     | 1.1 2.7 2.1 | COMPLETE | COMPLETE | COMPLETE |
| Exterior textile                  | 2.0 3.1 3.5 | COMPLETE | COMPLETE | COMPLETE |
| Interior textile                  | 0.8 1.4 1.3 | COMPLETE | COMPLETE | COMPLETE |
| Head harness                      | 0.6 0.8 0.7 | COMPLETE | COMPLETE | COMPLETE |
| Interior elastic                  | 1.2 1.5 1.3 | COMPLETE | COMPLETE | COMPLETE |

Figure 4. Opacity results.

| Table 2. Subjective appreciation results |
|-----------------------------------------|
| Subjective appreciation results         |
|-----------------------------------------|
| Prior to HALT                           | After 50 HALT               |
| The test item is in good condition      | The test item is in good condition |
| Plastic visor transparency: the background can be clearly seen through the visor | Plastic visor transparency: the background can be clearly seen through the visor |
| The elastic meets its function          | The elastic meets its function |
| All the materials are in good condition | All the materials are in good condition |
| All the materials present a uniform colour | Minor colour changes were identified in the Head Harness Textile, with the grey shade presenting a slightly lighter tone on some areas |

3.3.3. Quantitative test
3.3.3.2. Colour test. An important aspect of addressing the compatibility of PAPR with VHP is ensuring that colours do not change throughout the lifecycle of the item, since colour changes can be a sign of further physical and/or chemical alterations. For this study, colour measurements were taken on the different materials of the PAPR before and after exposure to the 50 HALT cycles, and such values were later used to calculate the colour difference between both stages. The results for the Colour Test performed on the different materials of the PAPR are shown in Figure 6.

4. Discussion

4.1. PAPR Decontamination using VHP

The purpose of decontamination technologies used in healthcare is to reduce the level of pathogenic microorganisms to a safe level. As a result, any new technology intended to be implemented in healthcare environments must show efficacy against harmful pathogens. Biological Indicators (BI) in the form of Geobacillus stearothermophilus spores were used to address the efficacy of Vaporised Hydrogen Peroxide (VHP) decontamination processes. VHP successfully killed all microorganisms present on the BI spores. Considering that Geobacillus stearothermophilus spores are challenging microorganisms to kill, the results obtained are promising and indicate that VHP has the potential to successfully kill other types of microorganisms.

Chemical Indicators (CI) were used to confirm that exposure to VHP took place at different challenging chamber locations. All CI used successfully showed a colour change from brown to green, indicating that VHP exposure was consistently delivered through different chamber locations. In addition, Decontamination Process Reports were created after each cycle, indicating if parameters required for the VHP technology to deliver a successful cycle were met. All Reports had a successful outcome, which not only includes the three decontamination cycles prior to the Off-Gassing Test, but also the 50 HALT Test Cycles.

Overall, the results from this study demonstrate that VHP is an effective and repeatable process for PAPR. In addition, the different efficacy and exposure controls implemented in this study (BI, CI, and Process Reports) can be easily implemented during real-life operations as a routine validation check of the technology.
4.2. PAPR off-gassing time
Any new decontamination technology intended to be implemented in healthcare not only has to be effective, but also must be safe to use. Decontamination methods that use chemical agents, such as VHP, should not leave harmful chemical residues, since they can lead to inhalation and/or dermal exposure risks (Kumkronga et al., 2021). While this is an important safety aspect, it unfortunately has not received the attention it deserves. Most studies on the effects of decontamination technologies on PPE focus on the efficiency and compatibility of such technologies, providing results for filtration efficiency, fit, and physical properties (Yim et al., 2020; Saini et al., 2020; Cramer et al., 2021; Al-Hadyan et al., 2021; Schwartz et al., 2020; Christie-Holmes et al., 2021). Limited studies have been found to analyse the chemical residues of decontamination technologies (Salter et al., 2010; Kumkronga et al., 2021; Viscusi et al., 2009; Radl et al., 2011); however, none of them include reusable PPE among the tested items.

As part of this study, an Off-Gassing procedure was developed to measure the parts per million (ppm) of Hydrogen Peroxide (HP) at different locations on the PAPR. The proposed method is non-destructive and therefore readings can be taken on the same item over time. The results from the Off-Gassing Test show that with the appropriate Off-Gassing Set Up, it is possible to off-gas one PAPR within 8 hours after decontamination. These results are considered significantly better than the ones obtained at the beginning of the preliminary phase, when it took more than 2 days for the PAPR to Off-Gas without using a dedicated set-up. Results also show that HP is desorbed differently depending on the material, observing longer desorption times on the Head Harness Textile and the Internal Elastic.

4.3. PAPR Compatibility with VHP
Any decontamination technology that is being considered for the decontamination of PPE must be compatible with PPE components and materials. Since most technologies were originally created for decontaminating environments and hard surfaces, there is limited evidence of the effects of exposure to VHP on soft surfaces. Most of the PPE compatibility studies emerged in response to the COVID-19 pandemic and focus on disposable items made mostly of nonwoven materials. While such studies greatly contribute to this research field, there is a need for additional evidence regarding reusable PPE items, which are mostly manufactured using woven materials.

To evaluate if the VHP decontamination technology would pose any negative effect on the material’s integrity, a PAPR was exposed to a Highly Accelerated Life Cycle Test (HALT), consisting of 50 decontamination cycles (a number that was considered to give a good estimation of the lifecycle of the product). A Visual Test was performed before and after the HALT test to address any possible changes (quantitatively and qualitatively) to the PAPR caused by the VHP technology.

The results from the Visual Appearance Test show that exposing a PAPR to 50 decontamination cycles did not lead to significant changes in the PAPR’s colour, opacity, or overall visual appearance. One of the areas of importance was the plastic visor, since a significant change in opacity could negatively impact the vision of the user. Results from the Photographic Appearance, Subjective, and Opacity tests confirmed that any possible changes on the item are not significant. Another important aspect was the colour of the item since changes in colour can be an indication of changes in the material. During the Photographic and Subject tests, it was possible to identify a minor colour change on the Head Harness Textile, an observation that was further confirmed with the colour difference value obtained on the Colour Test. While results show colour difference on all PAPR locations, with higher values on the Head Harness Textile and Internal Elastic, further testing and information would be...
required to identify the acceptance criteria for this PPE item and determine whether this might have an impact on the product’s performance.

**4.4. Conclusions**

Since the onset of the COVID-19 pandemic, there has been a marked increase in the demand for PPE to protect healthcare workers, first responders, and patients. The supply became insufficient and, as a result, decontamination of disposable PPE emerged as an alternative solution to extend the lifecycle of such items. Multiple studies have addressed the efficacy and compatibility of decontamination technologies with disposable PPE, with great results observed with VHP technologies. While most studies focused on disposable items usually made of nonwoven materials such as Filtering Facepiece Respirators, it is considered that such technologies could be also implemented to optimise the use of reusable items by providing an easier and more efficient way of decontaminating them.

This study evaluated the efficacy and compatibility of VHP as a decontamination method for PAPR reusable items. Special attention was paid to the Off-Gassing time required for hydrogen peroxide residues on the PAPR materials to reach undetectable levels (0 ppm), and to determine if exposing the PAPR to repeated decontamination cycles would negatively affect the visual properties of the PPE item.

Findings from this study showed that VHP is an effective and consistent method for the successful decontamination of PAPRs, using a commercially available decontamination system that takes less than 2 hours to complete. Off-gassing tests showed that 8 hours of off-gassing are required for the PAPR to be safe to reuse without becoming a health hazard. In terms of compatibility, findings showed that exposing a PAPR to 50 decontamination cycles does not result in significant changes in the PAPR’s colour, opacity, or overall visual appearance. Results from both the off-gassing and visual appearance tests indicate that the Head Harness Textile and Internal Elastic materials react differently to VHP, expressed through longer off-gassing times and larger colour differences.

The main advantage of this study is that it provides a real world practical solution for safely decontaminating PAPRs in healthcare systems equipped with VHP technologies. Not only were the research objectives developed as a response to healthcare needs, but the laboratory study was also performed using the same commercially available VHP technology and decontamination chambers that are used in real healthcare scenarios, allowing for a rapid implementation of the proposed decontamination approach. In addition, the efficacy of the VHP technology was thoroughly assessed by performing repeated testing using two indicators (BI and CI) in challenging chamber locations, which is considered another great advantage of the study.

Recent trends indicate an interest in increasing the use of reusable items for environmental reasons, but such items can be challenging to properly decontaminate and therefore introduce a risk of infection and/or cross-contamination. There is a need for safe and effective decontamination methods that are compatible with the materials commonly used in healthcare and that are ready to be implemented now. This study shows that VHP is a safe and effective method that is easy to implement and compatible with healthcare’s demand for fast processes and quick turnaround times.

The main limitation of this study is that the Off-Gassing Test was performed using 1 PAPR item, due to material constraints and limited resources during the pandemic. It is therefore unknown what the off-gassing values would be observed when testing the chamber at full capacity with
a maximum of 270 PAPR. However, due to limited studies performed on this research area, this study still provides an appropriate baseline for future testing to be performed with higher sample sizes as well as with a greater variety of PPE items.

To further advance this research area, off-gassing studies should be performed with decontamination chambers at full capacity in order to closely replicate real-life scenarios. In addition, as shown in this study, VHP can have different effects on different materials. It is therefore necessary that efficacy, off-gassing, and compatibility studies are performed at both product and sample level, to get an adequate understanding of how each material reacts so that this information might be used to create more compatible products.

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Notes
1. Vaporised Hydrogen Peroxide
2. Powered Air-Purifying Respirators
3. Highly Accelerated Life Cycle Test

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Supplementary material
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Appendix I
Decontamination using Vaporised Hydrogen Peroxide (VHP)

Vaporised Hydrogen Peroxide (VHP) Technology

The ProXcide™ decontamination system is a portable decontamination unit designed to fumigate enclosed spaces with Vaporised Hydrogen Peroxide (VHP) in order to decontaminate surfaces and items within those spaces. The system is fully automated and uses ultrasonic vapourisation technology to create microscopic droplets of hydrogen peroxide that are forced-evaporated to create hydrogen peroxide vapour. The ProXcide™ decontamination system holds a hydrogen peroxide bottle with a 7.5% Hydrogen Peroxide solution that fits into the main unit.

The ProXcide™ VHP system includes a Main Vapour Generating Unit placed inside a sealed room that controls the decontamination cycle parameters to ensure an effective and safe decontamination process; a Process Analyser/Electronic Indicator built into the main unit of the ProXcide™ that measures initial environmental parameters (e.g., temperature and relative humidity) to determine injection time and ensure a sufficient concentration of hydrogen peroxide is achieved and maintained; and a Process Monitor to control the Main Vapour Generating Unit from outside of the room being decontaminated.

The ProXcide™ system decontamination process involves the following steps:

1. Pre-Start Checks: the system checks functional and environmental parameters before enabling cycle preparation.

2. Start Cycle Preparation: the system prepares to start the decontamination process.

3. Vapour Diffusion (Injection): the system generates and circulates hydrogen peroxide vapour until required concentrations are achieved.

4. Surface Treatment (Dwell): the system maintains hydrogen peroxide concentration to ensure sufficient dwell time and treatment of all surfaces.

5. Deactivation: the system actively reduces the concentration of hydrogen peroxide vapour in the treatment space until a safe level is reached. ProXcide™ has a built-in deactivation module—catalysts to convert the hydrogen peroxide (H2O2) into water and oxygen (H2O & O2). Environmental levels must be reduced below safety limits of 1.0 ppm before the cycle can be considered complete.

6. System Reset: the system resets in readiness for the next process cycle.

Decontamination Chamber

The study was carried out using Inivos’ ProXpod decontamination chamber with the following dimensions: 2,400 mm × 2,400 mm × 2,100 mm (WxDxH) and door size 838 mm × 1,981 mm. ProXpods are fully sealed and have two doors with windows, door hinges, locks, and handles.

The ProXpod was fitted with four wire shelving units to help position the test items—two measured 1,200 mm × 600 mm × 1,800 mm (WxDxH) and the other two measured 1,000 mm × 600 mm × 1,800 mm (WxDxH). All four shelving units were fitted with five shelves. Each wire
shelving unit was assigned a number and each individual shelf was also given a number (e.g., the second shelf from the ground on unit 3 would be 3.2). The top, middle, and bottom locations were tagged with corresponding letters. These numbers and letters were used to define the Biological and Chemical Indicator locations.

**Decontamination Procedure**

For each Decontamination Cycle, the ProXcide™ was placed in the centre of the ProXpod chamber and the PAPR 01 was placed on the middle shelf between shelving units 1 and 3. Biological Indicators were placed in labelled sterile petri dishes and positioned inside the ProXpod with the lids open at the specified locations. Chemical Indicators were also placed at the specified locations using labelled tape. After placing all the test items, the ProXcide unit was operated as per the user manual to run a complete decontamination cycle.

Upon completion of the decontamination cycle, with the Process Monitor indicating successful completion of the cycle, the chamber door was slightly opened to measure the H2O2 gas level inside the room using a Porta Sens D16 III H2O2 gas monitor. The reading taken was required to be at 1 ppm or below for the room to be safe to enter into. Once it was safe to re-enter the chamber, the PAPR was taken to the Off-Gassing Test, and the Biological and Chemical Indicators were retrieved and processed.

**Appendix II**

Colour difference calculation

\[
\Delta E_0 = \left( \frac{\Delta L'}{k_c S_C} \right)^2 + \left( \frac{\Delta C'}{k_c S_C} \right)^2 + \left( \frac{\Delta H'}{k_c S_C} \right)^2 + R_f \left( \frac{\Delta C'}{k_c S_C} \frac{\Delta H'}{k_c S_C} \right)
\]

Where:

\( L_1^*, a_1^*, b_1^* = \text{colour coordinates from Colour Meter at 0 HALT} \)

\( L_2^*, a_2^*, b_2^* = \text{colour coordinates from Colour Meter at 50 HALT} \)

\[\Delta L' = L_2^* - L_1^*\]

\[\bar{L'} = \frac{L_1^* + L_2^*}{2}\]

\[\bar{C} = \frac{C_1 + C_2}{2}\]

\[a_1' = a_1 + \frac{a_2}{2} + \left( 1 - \frac{a_2}{2} \sqrt{\frac{a_2^2}{c_2^2 + 2}} \right), \quad a_2' = a_2 + \frac{a_1}{2} + \left( 1 - \frac{a_1}{2} \sqrt{\frac{a_1^2}{c_1^2 + 2}} \right)\]

\[C' = \frac{C_1 + C_2}{2}, \quad \Delta C' = C_2 - C_1, \quad \text{where} \quad C_1 = \sqrt{a_1^2 + b_1^2}, \quad C_2 = \sqrt{a_2^2 + b_2^2}\]

\[h_1' = \text{atan2}(b_1', a_1') \mod 360^\circ, \quad h_2' = \text{atan2}(b_2', a_2') \mod 360^\circ\]

\[
\Delta H' = \begin{cases} 
h_2' - h_1' & |h_1' - h_2'| \leq 180^\circ \\
360^\circ - h_2' + h_1' & |h_1' - h_2'| > 180^\circ
\end{cases}
\]

\[h_1' - h_2' \leq 0, \quad h_1' - h_2' > 0, \quad h_1' - h_2' \leq 0, \quad h_1' - h_2' > 0\]
\[
\Delta H' = 2 \sqrt{C_1 C_2 \sin(\Delta h'/2)},
\]
\[
H' = \begin{cases} 
\frac{(h_1' + h_2')}{2} & |h_1' - h_2'| \leq 180^\circ \\
\frac{(h_1' + h_2' + 360^\circ)}{2} & h_1' - h_2' > 180^\circ, h_1' + h_2' < 360^\circ \\
\frac{(h_1' + h_2' - 360^\circ)}{2} & h_1' - h_2' > 180^\circ, h_1' + h_2' \geq 360^\circ 
\end{cases}
\]
\[
T = 1 - 0.17 \cos\left(\bar{H} - 30^\circ\right) + 0.24 \cos\left(2\bar{H}\right) + 0.32 \cos\left(3\bar{H} + 6^\circ\right) - 0.20 \cos\left(4\bar{H} - 63^\circ\right)
\]
\[
S_L = 1 + \frac{0.015 (\ell - 50)^2}{\sqrt{20 + (\ell - 50)^2}}
\]
\[
S_C = 1 + 0.045 \ell
\]
\[
S_R = 1 + 0.015 \ell \bar{T}
\]
\[
R_T = -2 \sqrt{\frac{C_7}{C_7 + 25^2} \sin \left[ 60^\circ \exp \left( -\left( \frac{\bar{H} - 275}{25} \right)^2 \right) \right]}
\]
\[
K_L = 2
\]
