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Decontamination of a HEPA Filter Using Hydrogen Peroxide Vapor

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ABSTRACT

In the past, the life science and research community has decontaminated laboratories and lab equipment with highly toxic and carcinogenic chemicals such as formaldehyde and ethylene oxide. Since the EPA approved the use of hydrogen peroxide vapor (H_2O_2) for use in decontamination, the compound has become increasingly popular because it decomposes to oxygen and water, which are benign compared to the alternatives. This research demonstrates that H_2O_2 can be used to decontaminate HEPA filters, which pose a challenge because of their deep pleats that must be penetrated to ensure decontamination.

DECONTAMINATING WITH HYDROGEN PEROXIDE

Unlike other common sterilants, hydrogen peroxide is noncarcinogenic and non-mutagenic. The vapor breaks down into oxygen and water, which are both environmentally benign. Current theories hold that the oxygen radical released as the vapor decomposes is responsible for the highly lethal effect of the vapor on microorganisms. The compound has been shown to be effective against pathogenic bacteria, yeast, fungal spores and viruses.

Additionally, studies have also demonstrated that hydrogen peroxide vapor can penetrate and sterilize polypropylene, polyester, TYVEK and KIM-GUARD, and other materials often used in laboratories.

H_2O_2 does, however, have a few limitations. It can react with nylons and neoprenes, weakening them and reducing the life of parts made from these materials. Also, cellulosic materials such as wood or particle board can absorb the vapor. Absorption causes a problem after decontamination. The vapor is released slowly, which can cause air in enclosed spaces to exceed the 1 PPM threshold limit value established by the American Council of Governmental and Industrial Hygienists. Finally, H_2O_2 is not effective in sterilizing liquids. Otherwise, hydrogen peroxide has significant advantages over other sterilants, which explains its expanding role in decontamination.

One basic aspect of H_2O_2 behavior suggests that it may be difficult to use in decontaminating HEPA filters. The vapor decomposes very rapidly. To assure an effective kill rate, the gas must come into intimate contact with all surfaces which may harbor microorganisms before it decomposes to release the oxygen radical. The deep pleats of a HEPA filter are not easily penetrated by vapor moving by diffusion alone. This experiment was designed to avoid this potential problem by actively circulating H_2O_2 with a blower, which carries the vapor through the full depth of the filter.

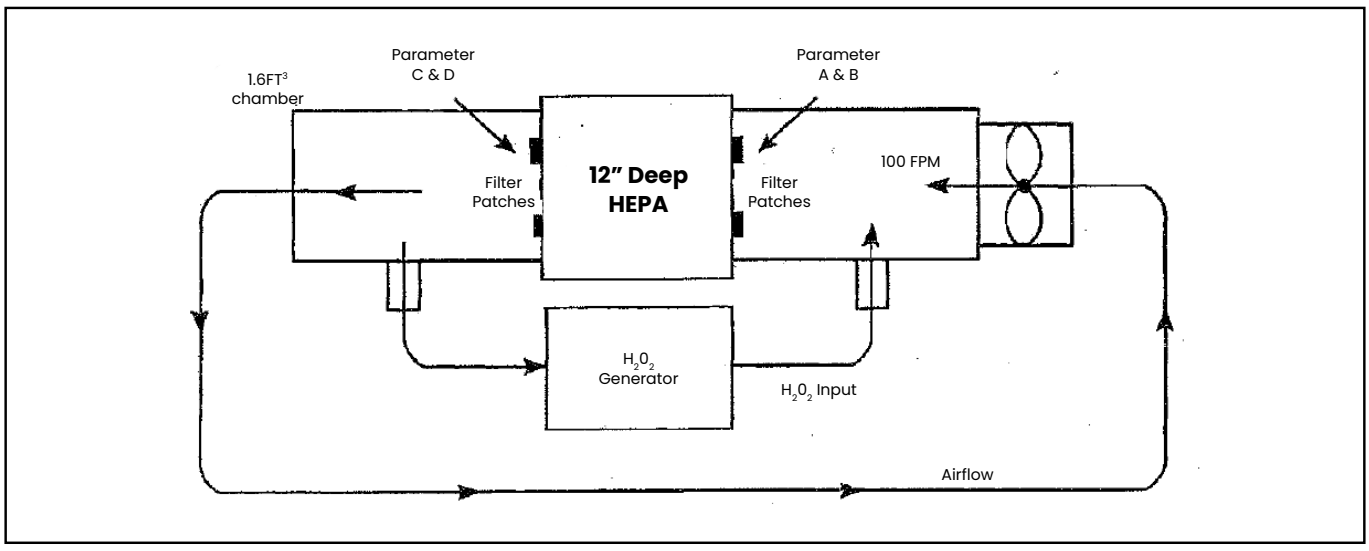

Figure 1. HEPA filter decontamination test assembly

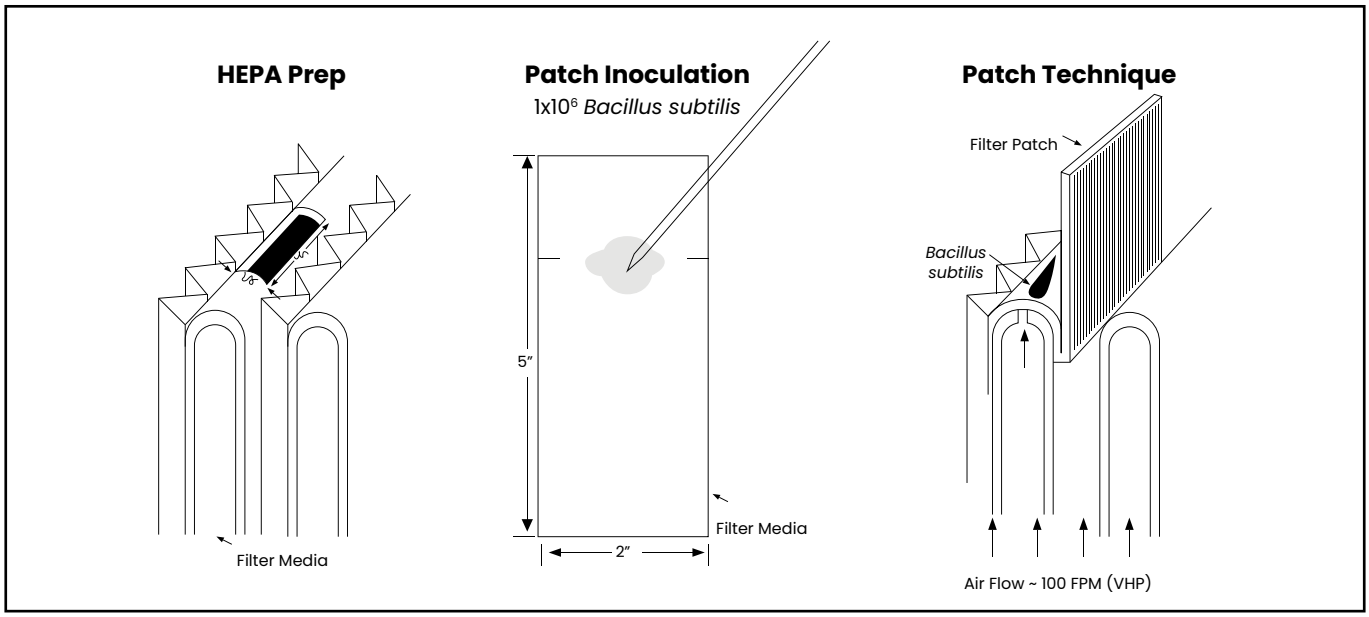

Figure 2. Preparation of the filter and the innoculated filter patches.

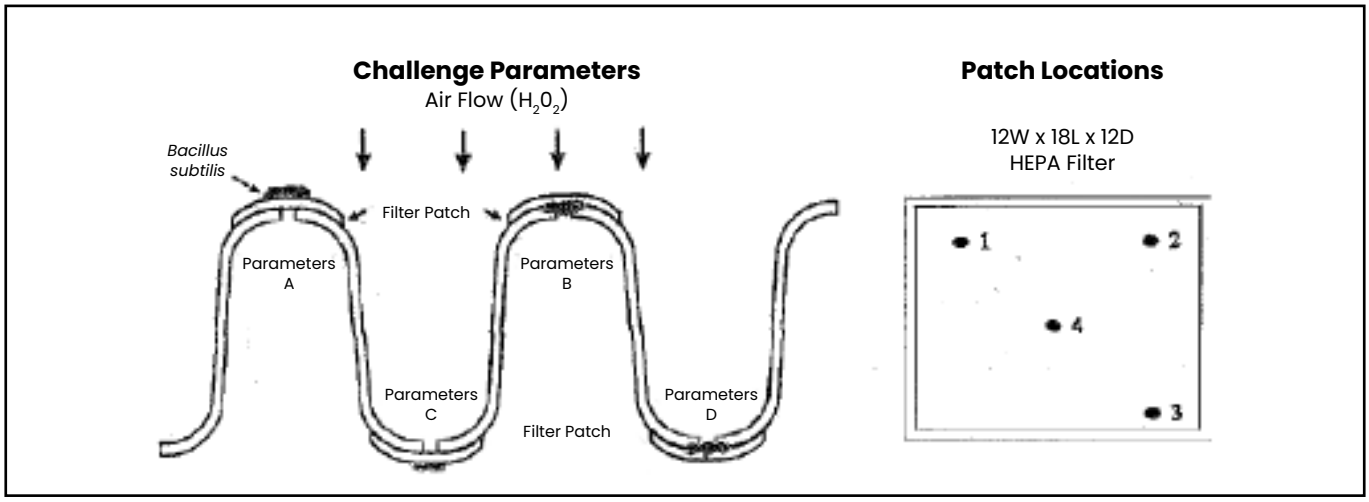

Figure 3. Patch locations and the challenge parameters.

RESEARCH PLAN & PROCEDURES

The objective of this research was to determine if hydrogen peroxide vapor could decontaminate effectively through the full depth of a HEPA filter. The equipment used included a 12" x 12" x 18" Donaldson filter and the AMSCO Model VHP™ 1000 generator. Spores of the Bacillus Subtilis bacterium were used as the contaminant, and these were impregnated into patches made from HEPA filter paper. The filter with contaminant patches was mounted in a chamber made from stainless steel and equipped with a blower assembly as shown in figure 1.

The first task consisted of impregnating the Bacillus Subtilis spores into the patches, which cannot be done with water because the material is hydrophobic.

Since alcohol is not sporicidal, and is readily absorbed by the media, the inoculum consisted of a 0.1 ml dose of 1 x 10^6 BG spores suspended in alcohol. Four patches were impregnated with the inoculum and mounted in three of the corners and in the center of the filter. To eliminate the possibility that decontamination depends on which side of the filter the contaminant is located, separate cycles were run with the inoculated side of the patches mounted "face-down" in the filter. To determine if H_2O_2 vapor is effective after travelling the full 12 inch depth of the filter, patches were mounted on the downstream side of the filter as well as the upstream side.

To remove the impregnated patches after exposure, they were configured with pull-tabs. Slots were cut in the original filter pleats, and the patches inserted over the slots as shown in figure 2. This was done to ensure that the air flow through the filter stayed relatively constant in spite of the extra layer of paper created by the patch.

We used the AMSCO cycle development guide to develop the cycle parameters including the air flow rate, vapor concentration and the length of exposure. After these were optimized, they were held constant through the tests at 12 cfm, 3.2 mg/l and 30 minutes, respectively.

Twelve decontamination cycles were completed; three at each of the four challenge parameters defined in figure 3. For disassembly after each test cycle, the apparatus was placed inside a horizontal-flow clean bench to reduce the risk of contaminating the inoculated patches as they were removed from the filter.

After removal, the patches were aseptically placed in trypticase soy broth and incubated at 37°C. Decontamination was considered successful if there was no visible growth of Bacillus Subtilis after seven days of incubation. To confirm that the inoculated patch supported growth before decontamination, an unexposed sample patch was also incubated for seven days as a control.

Patch Location

Challenge Parameter	1	2	3	4	Total
A	3/3	3/3	3/3	3/3	12/12
B	3/3	2/3	3/3	3/3	11/12
C	3/3	3/3	3/3	3/3	12/12
D	3/3	3/3	3/3	3/3	12/12
Total	12/12	11/12	12/12	12/12	47/48

No Growth / Trials

Table 1. Decontamination test results

RESEARCH PLAN & PROCEDURES

The results are shown in table 1. When filter patches inoculated with 1 x 10^6 Bacillus subtilis spores were exposed to 3.2 mg/l of H_2O_2 for over 30 minutes, all bacteria were killed in 47 of 48 tests.

The single patch showing growth had been placed at location 2 and tested under challenge parameter B. That is to say the patch was in one corner of the filter, the inoculation side of the patch was “down” with respect to the air flow direction and the patch was located on the upstream side of the filter (see figure 3). To consider this result in context, note that 23 of the 24 patches positioned in location 2 were successfully decontaminated, and of those, 2 of 3 patches were decontaminated under challenge parameter B.

These results indicate that patch location and orientation of the inoculation had little or no effect on decontamination. The single patch showing growth may have been contaminated between removal from the filter and the incubation process.

All of the control patches showed growth after the first day of incubation.

DISCUSSION

Successful decontamination of 47 of 48 test patches is a useful first step towards a procedure for decontaminating HEPA filters with hydrogen peroxide vapor. Additional studies will be required to ensure that H_2O_2 can successfully decontaminate HEPA filters from a normal lab environment, which may contain a broad range of microorganisms. While hydrogen peroxide vapor has been shown to successfully kill many types of bacteria, viruses and fungi under low organic loads, the addition of chemical loads may reduce or reduce its effectiveness.

This study has shown that when exposed to 3.2 mg/l of H_2O_2 for 30 minutes, a filter patch inoculated with 1 x 10_6 Bacillus Subtilis will be decontaminated. The tests do not indicate whether this success is a function of vapor concentration level, filter size, chamber material, exposure times, vapor distribution method or distribution uniformity. Many important questions remain. For example, if the chamber configuration were different, and if air were not moved actively through the filter, would these exposure times and concentration levels be sufficient for decontamination? The current study can be considered a useful first step towards answering the many complex questions which remain concerning filter decontamination.

CONCLUSION

Hydrogen peroxide vapor can decontaminate HEPA filter patches loaded with 1 x 10^6 Bacillus subtilis spores. Experimental limitations included clean HEPA filters, active circulation of H_2O_2 by a blower and the use of a controlled experimental environment.

REFERENCES

1. Rickloff, J.R., and Orleski, P.A. 1989. Resistance of Various Microorganisms to Vaporized Hydrogen Peroxide in a Prototype Tabletop Sterilizer. ASM meeting proceedings.

2. Klapes, N.A., Vesley, D. 1990. Vapor-phase Hydrogen Peroxide as a Surface Decontaminant and Sterilant. Applied and Experimental Microbiology, February 1990.

3. Klapes, N.A. 1990. New Applications of Chemical Germicides: Hydrogen Peroxide. ASM International Symposium.

4. Operating manual and cycle development guide for the Model VHPTM 1000 Generator. AMSCO, Apex, NC.

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