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Using Hydrogen Peroxide Vapor To Decontaminate Biological Safety Cabinets

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ABSTRACT

Recent research has shown that hydrogen peroxide vapor (H_2O_2) can be used to decontaminate biological safety cabinets (BSC's) as an alternative to using formaldehyde or ethylene oxide.^{1,2,3} H_2O_2 is non-carcinogenic, highly effective as a decontaminant and is environmentally benign. However, H_2O_2 vapor decomposes quickly, so the gas must be rapidly circulated throughout the BSC. Also, hydrogen peroxide vapor attacks some materials. Consequently, existing cabinets need physical changes and material substitutions so the potential advantages of H_2O_2 can be fully realized.

HYDROGEN PEROXIDE DECONTAMINATION COMPARED TO OTHER METHODS

Ethylene oxide gas and formaldehyde have traditionally been used to decontaminate BSC's. Both compounds are carcinogens. Their vapors must be neutralized before being vented to the atmosphere, and they can present hazards during clean up operations.⁴ H_2O_2 , in contrast, is not carcinogenic. The gas breaks down quickly into oxygen and water, and the decontamination procedure is a self-contained and automated process.

The exact decontamination reactions involving H_2O_2 are not well understood, but researchers hypothesize that the active component is the oxygen radical released as the H_2O_2 decomposes. Studies performed on a wide variety of fungi, bacteria and viruses show that D-values—which indicate the length of time needed to kill a specific concentration of organisms—are quite favorable with H_2O_2 .^{5,6}

DECONTAMINATION PROCEDURE

H_2O_2 decontamination requires four steps; drying the cabinet, concentrating the vapor, sterilizing the cabinet and finally diluting and removing any remaining vapor by ventilation.

Before sterilization, humidity inside the cabinet must be removed so the air can absorb H_2O_2 . In air, hydrogen peroxide vapor behaves much like water vapor (H_2O). Consequently, if the relative humidity is high inside the cabinet, the air will not have enough capacity to absorb the amount of hydrogen peroxide needed for effective decontamination. This is why a VHP™ 1000 generator begins its cycle by circulating dry air through the cabinet to remove existing water vapor.

After drying the cabinet, hydrogen peroxide is introduced at a high injection rate until the air is nearly saturated. At this concentration, organisms contacted by the vapor will be killed within two to three minutes based on D-values, but the vapor must be carried to all parts of the enclosure. Tests on typical cabinets have shown that the sterilization phase should last from 25 to 90 minutes, depending on the cabinet characteristics and on the uniformity of vapor distribution.

After full concentration is reached, the generator reduces the injection rate, but it continues to supply vapor to make up for the steady decomposition of H_2O_2 to oxygen and water. It is important to maintain a steady, nearly saturated level of vapor to ensure adequate decontamination—commonly defined as killing at least 1×10^6 organisms. After the sterilization phase is complete, the generator removes the air at a rate of 20 cubic feet per minute (cfm),

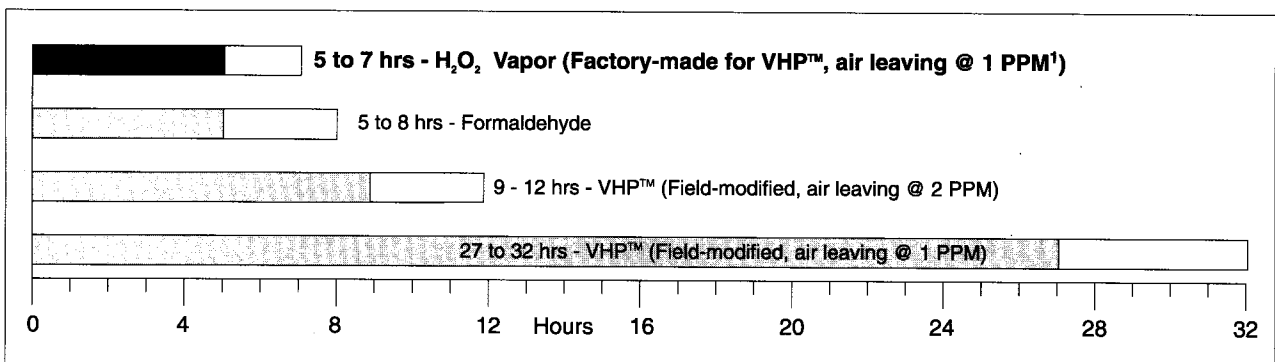


Figure 1. Comparing decontamination methods on the basis of hours required to kill 1×10^6 organisms

and breaks down any H_2O_2 it captures. Then the generator dehumidifies the air and returns it to the cabinet. This ventilation process continues until the vapor concentration falls to about 1 PPM.

VAPOR DISTRIBUTION

When formaldehyde or ethylene oxide are used for decontamination, the vapors are distributed partly by diffusion. Since these vapors are persistent, and since hours rather than minutes are needed for formaldehyde to kill enough organisms for full decontamination, this relatively slow distribution poses no problem. H_2O_2 vapor, in contrast, decomposes within a few minutes, which means there is not enough time for diffusion to carry the vapor to all parts of the cabinet. This is an important issue. When BSC's are sealed to isolate them for decontamination, there are "dead-end" plenums inside the enclosure where air does not circulate freely. To eliminate these dead spots and effectively decontaminate all surfaces, the H_2O_2 must be actively distributed throughout the enclosure. It is best to use the cabinet fan for this purpose.

While VHP™ 1000 generators use a fan to supply the vapor to the cabinet, that fan does not normally have enough capacity to accomplish uniform distribution. For example, a typical generator may circulate 20 cfm, compared to the cabinet fan which can move between 450 and 650 cfm. This mismatch of fan capacities creates a sharp concentration gradient between different sections of the cabinet. Unless specific provisions are made to supplement the generator circulation capacity, the sterilization process takes two or three times longer, a result which most users of BSC's would prefer to avoid.

Decontamination cycle time increases when vapor distribution is not uniform. In theory, if the H_2O_2 could be instantly and uniformly distributed throughout all corners of the cabinet, the sterilization phase would only require a few minutes—all surfaces and all organisms would be contacted simultaneously with vapor at full concentration, and very fast kill rates would be possible. Cabinet geometry, however, prevents instant, uniform vapor distribution, so more time is needed to build up full concentration in all corners of the enclosure. Decontamination time generally takes about an hour rather than a few minutes for this reason. Specific cabinet designs can either shorten or lengthen this time. For example, cabinets often have "dead-end" plenums with little air circulation so vapor cannot penetrate rapidly at full concentration. The easily-accessed part of such cabinets may be sterilized within a few minutes, but the dead-end plenums may not receive fully-concentrated vapor for much longer, because the H_2O_2 decomposes before it can reach into all corners.

Extended sterilization times lead to a second problem. In the long time it takes for vapor to reach into dead-end plenums, the more easily-accessed surfaces are exposed to full-strength H_2O_2 for a much longer period. Their surfaces may adsorb vapors which must be removed in the ventilation phase, extending that part of the decontamination cycle as well.

Consequently, vapor distribution uniformity and speed are important issues. For cabinets currently in use, modifications are necessary to achieve rapid, uniform distribution. Otherwise some of the theoretical improvements offered by H_2O_2 may not be achieved in practice, and total decontamination cycle time can be greatly extended.

MATERIAL COMPATIBILITY

Many materials are unaffected by hydrogen peroxide vapor, but others can absorb the gas and still others are attacked. For example, most HEPA filter frames are made of cellulosic material, which absorbs H_2O_2 at high concentrations, and then releases the gas slowly. Outgassing can continue for several days, depending on how the hydrogen peroxide reacts to the particular environment. This means the gas can build up—in low concentrations—in the work area and in the lab if the cabinet is not vented to the outside.

Nylons are an example of materials which are attacked by the vapor, and some nylons are used in electrical connectors and other electrical components. Similarly, some neoprenes are degraded by H_2O_2 , which means that existing gaskets may have to be replaced with a different material. A recent test of two gaskets suggests that neoprene may degrade in proportion to its exposed surface area. The dense gasket material lasted for many more decontamination cycles than an open-celled gasket made of similar material.

Breakdown of either electrical components or gaskets can be detrimental to safety, as well as adding maintenance expense and causing costly equipment down time. If the institution is considering the retrofit of existing equipment to use H_2O_2 vapor decontamination, cabinet materials should be identified and tested for compatibility.

Unfortunately, compatibility can only be established by exposing the material to hydrogen peroxide vapor. Just as liquid hydrogen peroxide cannot be used to predict sterilization performance of its vapor, liquid H_2O_2 cannot reliably predict degradation caused by the vapor nor the absorption behavior of materials when exposed to the gas.

DIFFICULTIES WITH H_2O_2 IN EXISTING CABINETS OR RETROFIT KITS

Existing cabinets can be modified in the field to accept hydrogen peroxide vapor generators, or standard cabinets provided with retrofit "kits". However, these alternatives are less than optimal because:

1. Such cabinets are not designed for fast distribution of H_2O_2 , and poor vapor distribution will require long cycle times—considerably longer than formaldehyde cycles.
2. Without material compatibility testing, important components may degrade unacceptably.
3. Unless cellulosic materials are replaced or treated they adsorb H_2O_2 , releasing it slowly during the ventilation phase. This slow release often explains why it may take 15 to 24 hours of ventilation to reduce the vapor concentration to 1 PPM.

RETROFIT ... Continued

4. Penetrations needed to connect VHP™ 1000 generators may void U.L., NSF or CSA certification.
5. It may be difficult to seal the cabinet to avoid vapor leaks, and informally-designed couplings may disconnect if bumped by custodial personnel or inexperienced workers who may not be fully aware of the potential hazards presented by H₂O₂ leaking into the lab environment.
6. If the cabinet blower is used to recirculate the air, there is the danger of inadvertently pressurizing the cabinet work area, causing plastic sheets and tape to blow off unexpectedly.

CABINETS SPECIFICALLY DESIGNED FOR HYDROGEN PEROXIDE VAPOR DECONTAMINATION

The end user may elect to purchase a cabinet specifically designed for H₂O₂ vapor decontamination. This alternative offers several potential advantages:

1. The manufacturer (rather than the end-user) is responsible for design changes which ensure successful results.
2. Eliminating dead spots and avoiding vapor adsorption by cellulosic materials achieves a cycle time of approximately 6 hours or less.
3. Changing H₂O₂-sensitive materials allows more cycles without degradation.
4. Replacing cellulosic materials avoids slow outgassing and the potential for vapor build-up in the work space and in the lab.
5. Carefully considered design details allow predictable cycle times and reliable procedures.
6. H₂O₂-related safety features are included in the cabinet design.
7. UL, NSF and CSA certification can be maintained.
8. Sealing mechanisms, integral to the cabinet design, are more effective and reliable than ad-hoc modifications.
9. The manufacturer provides documentation supporting the effectiveness of decontamination cycle times and procedures. This can be useful in validating a process and in assuring control of critical variables in research activities.

SUMMARY

H₂O₂ vapor decontamination offers safety and environmental improvements over formaldehyde. However, to achieve these advantages in existing equipment, extensive modifications are necessary to ensure that the gas is quickly and uniformly distributed to all parts of the cabinet. Also, it may be necessary to change some materials to avoid premature degradation and vapor absorption. New cabinets specifically designed for H₂O₂ decontamination are a better alternative, as they provide a number of useful benefits in addition to faster and more predictable decontamination cycle times than retrofitted cabinets.

REFERENCES

1. Jones, R., Large, S., Ghidoni, D., and Eagleson, D. 1991. *Decontamination of a Class II type B3 Biological Safety Cabinet using Vaporized Hydrogen Peroxide. American Industrial Hygiene Conference proceedings and ACUMEN, Vol.1., No.2; Baker Company, Sanford, ME.*
2. Jones, R., Large, S., Stuart, D., and Eagleson, D. 1991. *Sterilization of a HEPA Filter using Vaporized Hydrogen Peroxide. ABSA meeting proceedings and ACUMEN, Vol.1., No. 3; Baker Company, Sanford, ME.*
3. Klapes, N.A. 1990. *New Applications of Chemical Germicides: Hydrogen Peroxide. ASM International Symposium.*
4. U.S. Department of Health, Education, and Welfare. 1970. *Formaldehyde Decontamination of Laminar Flow Biological Safety Cabinets. National Institutes of Health.*
5. Rickloff, J.R., and Oreiski, P.A. 1989. *Resistance of Various Microorganisms to Vaporized Hydrogen Peroxide in a Prototype tabletop Sterilizer. ASM meeting proceedings.*
6. Toledo, R.T., Escher, F.E. and Ayres, J.C. 1973. *Sporicidal Properties of Hydrogen Peroxide against Food Spoilage Organisms. Appl. Microbiol. 26: 592-597.*

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