

EMERGING TECHNOLOGIES

The first dual-sterilant low-temperature sterilization system

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ABSTRACT

A new low-temperature sterilizer is described which uses both hydrogen peroxide and ozone in a multiphase process. The primary sterilant, vaporized hydrogen peroxide, is introduced into a chamber until a differential pressure of 19 torr is reached. By keeping the differential pressure constant, both the sterilant dose and exposure time are allowed to vary, which allows for a single cycle to be used to sterilize a wide variety of loads with differing size, material, and geometry. Lethality is achieved due to the effect of hydrogen peroxide in both the vapour and micro-condensation phases. Ozone is subsequently added to the chamber to decompose residual hydrogen peroxide and further increase lethality. Device performance has been validated by half-cycle, simulated-use, and in-use testing.

KEY WORDS

sterilization, hydrogen peroxide, ozone

INTRODUCTION

The STERIZONE® VP4 (VP4) Sterilizer is the first new low-temperature sterilization technology cleared by the U.S Food and Drug Administration (FDA) since introduction of the hydrogen peroxide gas plasma sterilizer in 1993 and the ozone sterilizer in 2003. The VP4 is also the first dual-sterilant device cleared by FDA for terminal sterilization of cleaned, rinsed, and dried metal and non-metal reusable medical devices.

The VP4 uses both vaporized hydrogen peroxide (VHP) and ozone in a multiphase process, providing a minimum Sterility Assurance Level of 10⁻⁶ (1). The sterilization cycle is compatible with a variety of materials and device geometries including general instruments, single channel flexible endoscopes, and rigid channel devices. The device can also sterilize up to 75 pounds of medical instruments in a single load (2). Device performance has been validated by not only half-cycle testing, but also simulated-use and in-use (within a hospital) testing.

Although both hydrogen peroxide (H₂O₂) and ozone are well-known sterilants, the process by which both are introduced, controlled, and combined within the VP4 sterilization chamber is unique. This results in different process parameters and chemistries, in comparison with first-generation sterilization processes. This paper describes the critical parameters, chemistry, and enhanced lethality found with the VP4 Sterilizer.

Sterilizer cycle description

The STERIZONE® VP4 Sterilizer is a self-contained stand-alone device, using VHP and ozone in a multiphase process. Unlike other low-temperature sterilizers, which require use of

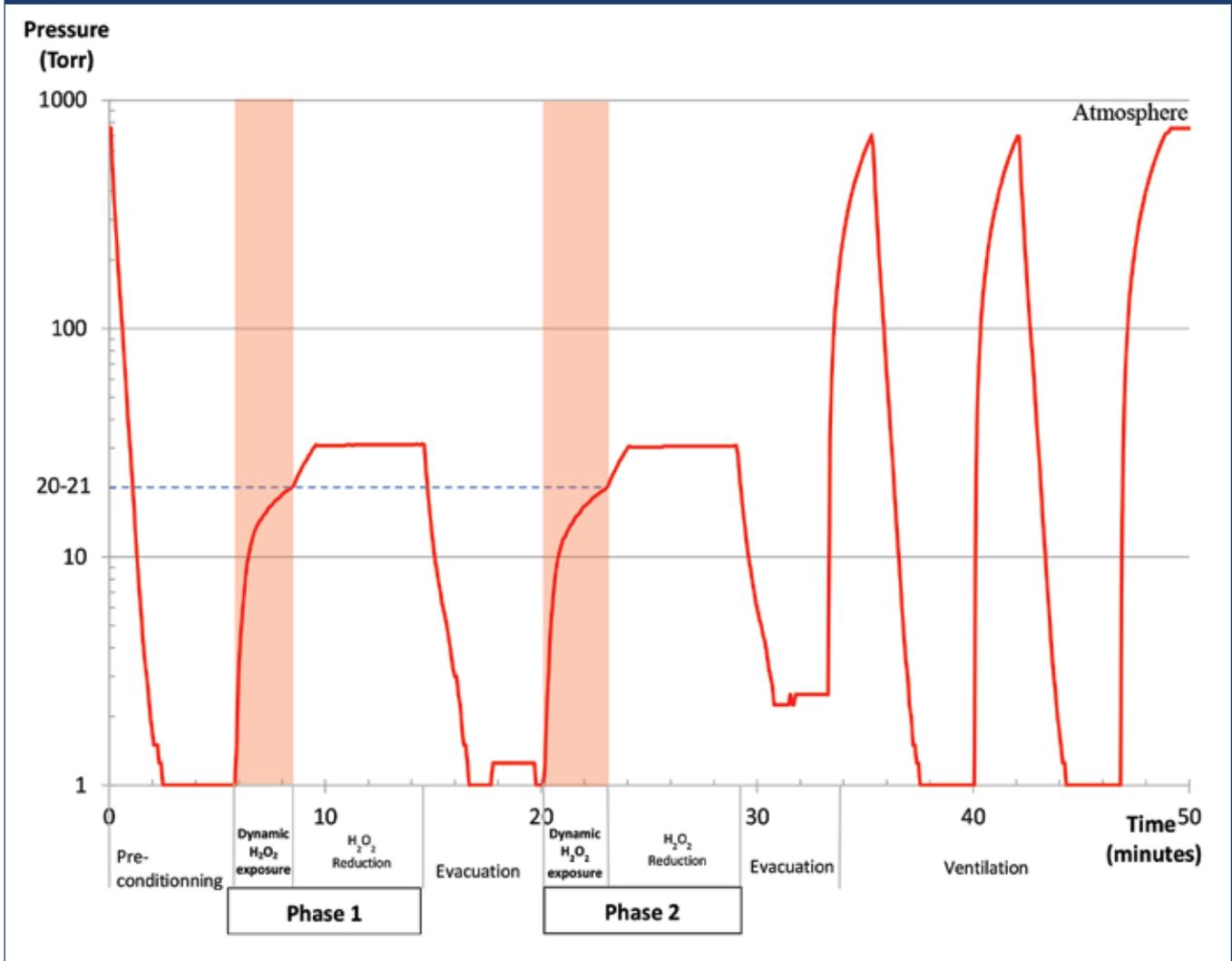
various cycles for different types of devices, the VP4 offers a single sterilization cycle ("Cycle 1") intended for all allowed substrates and geometries, including general instruments, single channel flexible endoscopes, and rigid channel devices. The process pressure and time profile for Cycle 1 is provided in Figure 1.

Upon loading medical devices into the sterilization chamber and closure of the door, the chamber is subjected to a vacuum of 1 torr (referred to as Pre-conditioning step). The Pre-conditioning step has a total maximum duration of 10 minutes, and is reconfirmed immediately following the degassing period.

The first cycle phase (Phase 1) is initiated with the Dynamic H₂O₂ exposure step. During this step, a 50 weight-percent H₂O₂ solution is injected in vapour form into the sterilization chamber until a differential pressure set point of 19 torr is reached (i.e., the actual chamber pressure is 20 torr, less the initial vacuum of 1 torr, which is equivalent to a "differential pressure" or "DP" of 19 torr).

Hydrogen peroxide vapour is generated within the VP4 by flash vaporization, meaning that the mixture of H₂O₂ and water vapour injected into the sterilization chamber is substantially the same weight percent composition as the multi-component liquid. Flash vaporization is achieved at elevated temperatures, which accounts for the respective boiling points of the components (H₂O = 100°C; H₂O₂ = 150.2°C; 50% aqueous solution of H₂O₂ = 114°C).

The VP4 incorporates a "Dynamic Sterilant Delivery System™", which provides continuous exposure to hydrogen peroxide through multiple small-pulsed injections of the sterilant (≈ 40 mg/pulse), with one pulse injected per second.

Figure 1: Cycle process pressure profile for Cycle 1 of the STERIZONE® VP4 Sterilizer.

The amount of sterilant introduced into the sterilization chamber is dependent upon reaching the set differential chamber pressure of 19 torr. This in turn means that both the total dose and time of sterilant exposure vary depending on the weight and composition of the load, and the load temperature (i.e., variables that effect differential pressure). This differs from first generation VHP devices, which employ static dose and time parameters (but variable chamber pressure). By keeping the differential pressure within the sterilization chamber constant at 19 torr, while allowing the dose and exposure time to vary, a single cycle can be used to sterilize a wide variety of loads with differing size, material, and geometry.

The second step of the cycle phase is the H₂O₂ reduction step. During this step, 2 mg/L of ozone is injected into the chamber, followed by a five-minute dwell time. This step is intended to reduce residual hydrogen peroxide, which may have been preferentially absorbed by certain polymers. This step also enhances microbicidal efficacy via the formation of hydroxyl

radicals. However, a 6-log spore reduction is consistently achieved for all reusable devices that fall within the sterilization claims after exposure to only the first Dynamic H₂O₂ exposure.

During the second cycle phase (Phase 2), the same sequence is repeated, including the Dynamic H₂O₂ exposure and H₂O₂ reduction steps. The full cycle is then completed with an evacuation and ventilation, through a catalytic converter, which decomposes excess hydrogen peroxide vapour into water and oxygen. Since the sterilization chamber remains sealed during all process steps, there is no occupational or environmental exposure to sterilants.

Because differential pressure is the critical process parameter for the VP4 Sterilizer, and not dwell time or sterilant dose, both are allowed to vary depending on the load size, temperature and composition. This in turn allows for total cycle time to vary between 46-60 minutes, reflecting a variable Dynamic H₂O₂ exposure time of between 210-600 seconds (per half-cycle).

Micro-condensation of hydrogen peroxide vapour

VHP is, by definition, a gas when introduced into a chamber, reflecting the same weight percent composition as found in the prevaporized liquid solution. At typical room temperatures and atmospheric pressure, both water and H₂O₂ are predominantly liquid, with the headspace air within a closed container having a small amount of gas phase H₂O₂/H₂O that is in equilibrium with the liquid.

When VHP is initially injected into a chamber under vacuum, both H₂O₂ and H₂O remain in the gas phase. However, as the H₂O₂/H₂O gas concentration increases, coupled with encountering the cooler temperatures of the sterilization load, the H₂O₂/H₂O vapour will begin to condense into a microscopic layer, also known as the “micro-condensation” layer (3). The exact temperature required to condense moist, H₂O₂-laden gas is called the dew point (4). In a fixed temperature environment, dew point may also be expressed in terms of the pressure (or concentration) of H₂O₂-laden gas required for condensation (i.e., “dew pressure”). The relationship between dew point and condensation in a sterilization chamber is identical to the formation of fog in a moist environment (i.e., the dew point is the temperature at which the air becomes 100% saturated with water vapour, which condenses into water droplets, which we see as fog).

Once the dew pressure has been reached for a given temperature and weight fraction of H₂O₂, a micro-condensation layer (measured in micrometers, and thus not visible to the human eye) forms on the surface of the sterilization load, which is in equilibrium with the H₂O₂/H₂O vapour. However, because hydrogen peroxide has a lower equilibrium vapour pressure (i.e., lower dew pressure) versus water, it will preferentially condense into the micro-condensation layer. This in turn means that once the dew pressure has been reached, the equilibrium concentrations of H₂O₂ will be much higher in the liquid phase (>70%) versus the vapour phase (<10%), even for low weight-percent H₂O₂ solutions (5). The high concentration of H₂O₂ in the liquid phase is believed to be responsible for very rapid kill, which is greater than the corresponding gas-phase lethality, particularly for a low-temperature environment (6).

The extent of condensation for a given H₂O₂/H₂O weight percent depends on, among other things, temperature (both chamber and load). Figure 2 presents the theoretical dew pressure curves of a 50 weight-percent H₂O₂ solution at three different temperatures: 20°C, 30°C, and 40°C. As expected, the higher the temperature, the higher the dew pressure (i.e., the pressure required for the first condensate to form). Thus, at 20°C, the dew pressure is only 3.5 torr whereas at 40°C, the dew pressure is 13 torr.

Below the dew pressure, H₂O₂/H₂O is in the vapour phase. Above the dew pressure, a micro-condensation layer is formed on any exposed surface, with an equilibrium established between the gas and liquid phases.

Figure 2: H₂O₂/H₂O system theoretical dew pressure curves at 20°C, 30°C and 40°C (7).

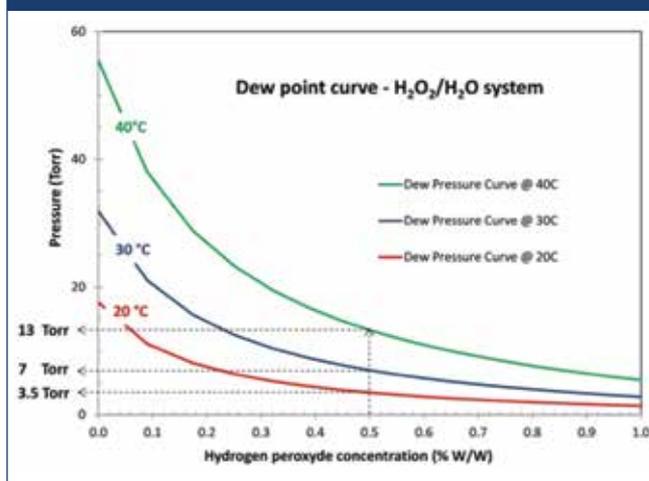
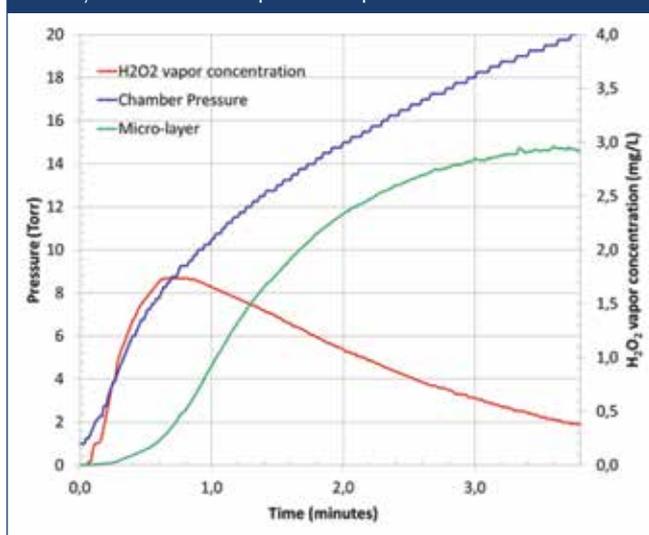


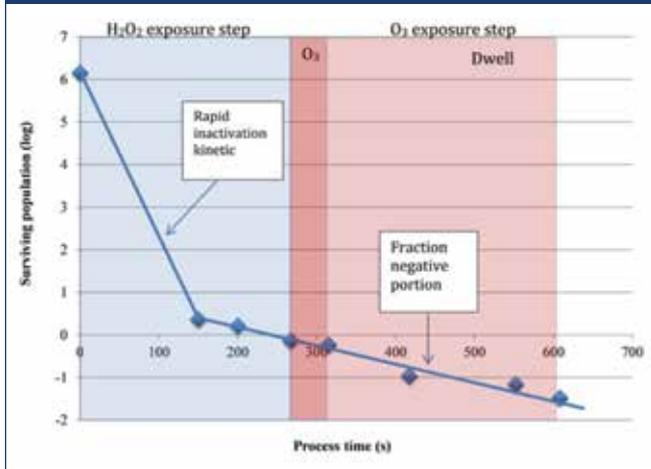
Figure 3: Condensation of the 125-280 Solution™ during the Dynamic H₂O₂ exposure step for a 50 lb load at 26°C.



In other words, as the pressure of vaporized H₂O₂/H₂O increases within a chamber, lethality is achieved due to the effect of both H₂O₂ in the vapour and micro-condensation phases.

Empirical data confirms the formation of a micro-condensation layer in the VP4, as detailed in Figure 3. Three variables were measured in the sterilization chamber during a standard hydrogen peroxide injection: chamber pressure (blue curve, expressed in torr), H₂O₂ vapour concentration (red curve, expressed in mg/L), and thickness of the micro-condensation layer (green curve, expressed in kÅ). Hydrogen peroxide vapour concentration was measured using UV spectroscopy whereas the thickness of the microlayer was measured using a crystal microbalance. The experiment was conducted at the upper end of the recommended load temperature for the VP4, namely 26°C, which is less likely to form micro-condensation.

Figure 4: Inactivation of the Test Pack on the rack only at $23^{\circ}\text{C} \pm 2^{\circ}\text{C}$, in the H₂O₂ exposure step followed by the O₃ exposure step (O₃ injection + dwell).



In the early phase of H₂O₂ injection, chamber pressure increases (blue curve), and H₂O₂ vapour concentration increases (red curve), without any meaningful change in the thickness of the micro-condensation layer (green curve). However, at approximately 30-40 seconds, the rate of change in micro-condensation layer increases, corresponding to an approximate peak in H₂O₂ vapour concentration. This point also corresponds to a chamber pressure (or dew pressure) of approximately 7-8 torr, which as discussed in Figure 2, is the pressure at which condensation begins to form. Thus, the experimental dew pressure is consistent with the theoretical dew pressure.

Once micro-condensation begins to form in the chamber, H₂O₂ vapour concentration drops in spite of the fact that H₂O₂ injection continues until the chamber pressure reaches 20 torr. The fact that the H₂O₂ vapour concentration decreases while the micro-condensation layer increases, confirms that micro-condensation is occurring within the chamber. If the injected VHP remained in the gas phase, the H₂O₂ vapour concentration would continue to increase over the complete injection cycle (corresponding to the increase in VHP over time), reaching the same concentration as the initial solution. Furthermore, the micro-condensation layer would remain minimal.

Thus, the VP4 achieves sterilization efficacy by use of vaporized hydrogen peroxide, which exhibits lethality in both the vapour and micro-condensation phases. By maintaining a constant differential pressure of 19 torr, a minimum micro-condensation layer is formed on all surfaces, which ensures lethality. Although the role of micro-condensation in conventional VHP sterilizers remains controversial (some manufacturers of conventional VHP sterilizers claim that the sterilant is always in the vapour phase), it is likely that all VHP devices form micro-condensation layers, particularly with sterilization loads processed at room temperature (8, 9). However, it is also likely that they are uneven and uncontrolled,

meaning that biocidal activity is primarily due only to hydrogen peroxide vapour, which is less efficient than microcondensation (10).

Differential pressure as a primary process parameter

The critical process parameters for controlling the formation of micro-condensation within a sterilization chamber include the differential pressure and load temperature. Both thermodynamic calculations and experimental data confirm that increasing the injection of H₂O₂/H₂O vapour beyond the dew point pressure (at a given temperature) will result in micro-condensate formation. However, increasing the DP to a specific target beyond the dew point pressure, is a function of the volume of H₂O₂/H₂O vapour injected into the system, the size and surface area of the sterilization load, and load temperature.

Experimental data confirms that a differential pressure of 19 torr will consistently sterilize the most challenging instruments/loads, at the highest allowed temperatures. This has been validated in half-cycle, simulated-use (where the most resistant microorganism to the sterilization process is mixed with organic and inorganic soils and inoculated onto devices) and in-use testing (medical devices soiled during actual hospital procedures are tested for sterility).

The size of the load (defined as weight and/or surface area) can influence the time required to reach the differential pressure since large loads allow for more micro-condensation. When H₂O₂/H₂O vapour is being condensed on large surface areas, a greater volume of vapour is required in order to maintain or increase the overall vapour pressure. Thus, the time required to reach a DP=19 torr in a large load will be longer than in a small load, since the Dynamic H₂O₂ delivery system operates at a fixed injection rate.

In addition to DP, experimental data has been generated to confirm that the load temperature should be between 20-26°C. As previously discussed, temperature plays an important role in determining the dew pressure, with increasing temperatures resulting in higher dew pressures. Although load temperatures above 26°C form a micro-condensation layer, experimental data shows that for certain types of instruments, sterilization efficacy is reduced above 26°C. This in turn is attributed to lower condensation levels and lower H₂O₂ exposure times (i.e., for a given load, the time required to reach a DP=19 torr is shorter at high temperatures). Since the VP4 uses continuous small-pulsed injections of H₂O₂ vapour until the differential pressure is reached, the total sterilant exposure time is limited to the time required to reach the differential pressure. If less time is required to reach differential pressure due to less condensation, the load has a lower exposure time to the sterilant.

Load temperature should not be confused with chamber wall temperature, which is set at 41°C in

the VP4. First generation VHP devices also set chamber wall temperatures at relatively high levels ($\pm 50^{\circ}\text{C}$), which discourages formation of micro-condensation on chamber walls. However, since sterilization loads are usually conditioned at room temperature ($\pm 23^{\circ}\text{C}$), load temperatures are much lower than chamber wall temperatures. This in turn has a direct effect on the formation of micro-condensation layers. Nonetheless, existing VHP devices do not provide load temperature restrictions, even though load temperature is crucial to maintaining a “dry” process (8, 9).

As noted above, load temperature and injection time are correlated in the VP4. This is because the device continuously injects H₂O₂/H₂O vapour into the sterilization chamber until the differential pressure of 19 torr is reached. However, at high temperatures ($> 26^{\circ}\text{C}$), it takes less time to reach 19 torr, particularly for small loads. Thus, by restricting the range of injection times, one can effectively account for load temperatures outside the range of optimum efficacy. Specifically, experimental data confirms that when the injection time is limited to between 210-600 seconds, “worst-case” sterilization loads warmer than 26°C are aborted (i.e., the minimum injection time of 210 seconds is not reached). Likewise, “cold” loads (under 20°C) are also aborted, since they exceed the upper limit of allowed injection time.

The foregoing discussion highlights a difference between the VP4 and first generation VHP devices. Whereas the VP4 maintains a constant differential pressure of 19 torr, with a variable dose and exposure time, conventional VHP devices allow for DP to vary, although dose and exposure time are kept constant. In addition, VHP devices do not control or address the issue of load temperatures, even though chamber and load temperatures are in practice very different. Finally, the VHP devices address the static nature of their process by incorporating multiple cycles into a single device, each targeting different loads, with differing VHP exposure times and dose.

Role of ozone

Within each cycle phase, after achieving a differential pressure of 19 torr, 2 mg/L of ozone is injected into the sterilization chamber followed by a five-minute dwell time. After injection of ozone, chamber pressure increases to 27-30 torr.

The primary purpose for this step is to reduce residual hydrogen peroxide, which may be preferentially absorbed by certain polymers (e.g., polyoxymethylene and polyurethane) (11). Residual hydrogen peroxide can render a material cytotoxic unless removed by secondary reaction or extended aeration.

Adding ozone to hydrogen peroxide also enhances overall microbicidal efficacy. The chemical reaction between ozone and hydrogen peroxide is known as the “peroxone oxidation” (12). In a typical application (e.g., water treatment facility), gaseous ozone is injected into a liquid containing

hydrogen peroxide with various contaminants. Contaminants are oxidized near the gas-liquid interface.

Experimental data confirms that ozone reduces residual hydrogen peroxide concentration in polymers with a high propensity towards absorbing VHP. For example, polyurethane has a 17% reduction in residual H₂O₂ when exposed to the H₂O₂ reduction step. Both in vitro and in vivo biocompatibility testing on material samples processed with the VP4 confirm that all common metallic and polymeric materials are non-toxic and safe for use.

Although a 6-log spore reduction is consistently achieved during the first Dynamic H₂O₂ exposure step, data confirms that the addition of ozone results in additional lethality. Using a specially designed Test Pack (which FDA required to have equivalent or greater resistance than worst-case devices and loads), the inactivation potential of the H₂O₂ reduction step was measured using process time (i.e., the only common variable between the hydrogen peroxide step as controlled by differential pressure, and ozone injection controlled by dose and dwell time). As shown in Figure 4, the inactivation profile is biphasic with the Dynamic H₂O₂ exposure step adding up to 1.8 log lethality, beyond the 6-log half-cycle reduction achieved from exposure to only VHP. Replacing ozone with oxygen resulted in minimal additional lethality, confirming that the reaction of ozone with H₂O₂ is responsible for the additional microbial potential.

Preliminary studies have confirmed that this additional lethality can be used to sterilize very challenging devices such as flexible colonoscopes, which currently are reprocessed using only high-level disinfection.

Finally, material compatibility is not compromised by the addition of ozone, which is known to be highly corrosive to certain materials used in medical devices (13). Because ozone preferentially reacts with residual hydrogen peroxide, it does not directly oxidize material surfaces. Thus, overall material compatibility of the VP4 process is comparable to conventional VHP sterilizers, in spite of the addition of ozone.

SUMMARY/CONCLUSIONS

The STERIZONE® VP4 (VP4) Sterilizer is the first new low-temperature sterilizer to be controlled by differential chamber pressure. Unlike conventional VHP devices, which maintain a constant dose and exposure time, but allow chamber pressure to vary, the VP4 maintains a constant chamber pressure, while allowing dose and time to vary depending on the load size and composition. This results in a single sterilization cycle able to process widely differing devices and weight without the need to select a preferred cycle.

Like first-generation low-temperature sterilizers, the VP4 achieves sterilization by use of vaporized hydrogen peroxide, which is an oxidizing agent known for its bactericidal, virucidal, sporocidal and fungicidal properties. However, lethality is based on both the vapour and micro-condensation forms of hydrogen peroxide, with the latter being recognized as having superior microbial kill rates.

A hydrogen peroxide reduction step has been added to Cycle 1 to reduce residual H₂O₂ preferentially adsorbed by certain polymers. Experimental data has been generated to prove that ozone reduces residuals in select polymers, but also results in additional lethality.

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