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### Effectiveness of Dry Hydrogen Peroxide on Reducing Environmental Microbial Bioburden Risk in a Pediatric Oncology Intensive Care Unit

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**Background:** Routine manual cleaning and disinfection of the health care environment is often suboptimal. Residual contamination poses an infection risk, particularly for immunocompromised patients. This study evaluates the efficacy of dry hydrogen peroxide (DHP) on microbial surface contamination in a pediatric oncology intensive care unit.

**Methods:** Surface samples from 5 high-touch and 2 low-touch surfaces were obtained for culture and adenosine triphosphate readings after manual cleaning on multiple days in 4 intensive care unit rooms, before and after DHP was deployed. Air samples were collected as well at the study site. Data outcomes were measured in terms of total colony-forming units for the cultures and relative light units for adenosine triphosphate.

**Results:** The overall mean surface microbial burden was significantly reduced in the intervention group compared to the control group (mean 5.50 vs 11.77,  $P < .001$ ). These reductions in colony forming units were seen across all sampling sites in the intervention group. A reduction in the mean relative light units levels was also noted in the intervention group when compared to the control group (172.08 vs 225.83,  $P < .006$ ). Reductions with the air samples were also noted ( $P = .139$ ).

**Conclusions:** Study demonstrates that DHP was effective in reducing microbial surface contamination and improves quality of environmental cleaning.

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## Introduction

The immunocompromised condition of children with cancer places them at high risk of infection acquisition.<sup>1</sup> Bacteria account for a considerable percentage of these infections and are associated with substantial morbidity and mortality.<sup>1</sup> The impact of the health care environment on horizontal transmission of bacteria and other microorganisms has been recognized as a major infection prevention and control priority in this population.<sup>2</sup> A clean environment is a critical component in an effective infection prevention program. A large body of evidence has demonstrated the extent to which the hospital environment can be contaminated with potentially pathogenic microorganisms.<sup>3-5</sup> These organisms can survive for extended periods of time in the environment,<sup>5-8</sup> and the risk for colonization or infection acquisition from the environment via either direct contact or indirect contact through health care workers' hands has been well described.<sup>9-11</sup>

The manual process of cleaning and disinfection are a necessary part of maintaining a clean and safe environment; however, a number of studies have demonstrated that the manual processes alone are suboptimal.<sup>9,12,13</sup> Additionally, research has shown that not only does manual cleaning not fully eradicate clinically important microorganisms such as methicillin-resistant *Staphylococcus aureus* (MRSA), vancomycin-resistant *Enterococcus*, multidrug-resistant *Acinetobacter*, and *Clostridioides difficile* from hospital environments, but also that those microorganisms represent a significant infection risk to the patients who subsequently occupy those spaces.<sup>12-16</sup>

Automated "no-touch" technologies are increasingly being employed to augment manual cleaning and disinfection in an effort to achieve more comprehensive environmental bioburden reductions.<sup>3</sup> While some studies have demonstrated effective microbial reductions with their use, many of these technologies are limited by logistical constraints.<sup>2,3</sup> These include operation in an unoccupied spaces due to safety concerns, staffing costs for operation, and efficacy parameters such as device placement and run-time.<sup>2,3</sup> Additionally, many of these technologies—much like manual cleaning—do not address the active, ongoing recontamination of the environment that inevitably occurs from patients, visitors, and staff alike.<sup>2</sup>

The present study evaluates the effectiveness of an automated microbial reduction technology that does not share the constraints or limitations associated with other no-touch technologies. The technology used in this study catalytically produces dry hydrogen peroxide (DHP) from ambient humidity and oxygen. DHP is delivered continuously throughout a space regardless of occupancy status. Hydrogen peroxide is a well-established disinfectant with a strong safety profile in the pediatric population.<sup>17,18</sup> Research has shown airborne hydrogen peroxide, in particular, to be an effective disinfectant for the hospital inanimate environment,<sup>17</sup> but its efficacy has historically been counterbalanced by its restriction for use in unoccupied spaces.<sup>3</sup> DHP possesses the broad-spectrum antimicrobial activity of hydrogen peroxide. It exists in a nonaqueous, gaseous state. DHP is used at concentrations far below acceptable safety limits for human exposure established by the Occupational Safety and Health Administration (OSHA) and is safe for use in occupied settings.<sup>19,20</sup> Previous research has shown DHP to be highly effective in reducing bioburden in the hospital setting.<sup>21</sup> This study assessed the efficacy of DHP, used as an adjunct to standard manual cleaning, in reducing microbial contamination in the air and on surfaces within the intensive care unit of a pediatric oncology hospital in Guatemala.



## Methods

This prospective cohort study was performed over a month at the Pediatric Intensive Care Unit (PICU) at Unidad Nacional de Oncología Pediátrica (UNOP), a 65-bed pediatric oncology hospital located in Guatemala City, Guatemala. The unit has 9 individual private room beds with a centralized air conditioning system. No alterations in clinical activities or existing engineering controls were made throughout the study period. The study was approved by the facility's Institutional Review Board and Ethics committee. The use of DHP was designed to be an adjunctive microbial reduction strategy to the facility's standard cleaning and disinfection protocol.

### *Selection and Sampling*

Two rooms in the PICU served as controls and 2 rooms served as intervention DHP sites. Five high touch-sampling surfaces (bed handrail, bedside table, monitor, inside door handle, and nurse table) and 2 low touch-sampling surfaces (side of over-bed light and paper dispensers) swabbed for cultures and adenosine triphosphate (ATP) assay. We identified the swab surfaces using a visual sticker with a 2 x 2 inch swab surface area just below the sticker. During the preintervention phase, we obtained baseline surface culture and ATP swabs from the study areas on 3 separate days (Monday, Wednesday, and Sunday) during the week prior to deployment of portable DHP units in the intervention rooms. After deployment of DHP in the intervention rooms, we collected samples for surface culture and ATP from the same sites and days the following week then weekly for the next 4 weeks. We obtained the samples collected at the same time of day and after daily environmental cleaning and disinfection.

In addition to environmental surface sampling, we also collected air samples at baseline and during the intervention phase following the deployment of DHP. The air samples were taken weekly using settling plates for 15 minutes in each of the study rooms, hallway, and exterior samples taken just outside the hospital.

### *DHP Deployment*

The intervention phase of the study involved the deployment of one portable DHP Omnia Sentry stand-alone microbial reduction system plugged into a standard 120 V/50-60 Hz outlet (Synexis, Overland Park, KS) in each of the 2 intervention rooms. Each unit utilizes ambient oxygen and humidity in the room's air to catalytically generate DHP per manufacturer's instructions. The unit generates DHP around a concentration that ranges between 5 and 25 parts per billion (ppb),<sup>22</sup> well below the human safety exposure limit of 1-ppm long established by OSHA and more recently affirmed in a longitudinal study.<sup>20</sup> Unlike hydrogen peroxide mists or vapors, DHP is a true gas and is not generated from an aqueous solution. DHP behaves like oxygen and nitrogen, diffusing through the air to achieve a dilute equilibrium concentration. The units operated continuously—24 hours per day/7 days per week—in the occupied rooms throughout the intervention phase of the study.



### *Microbiology*

Specimens were cultured in Agar Plate Count (Merck®) for mesophile aerobic bacteria and in Potato Dextrose Agar (Merck) for yeasts and fungi incubated at 35  $\pm$  0.5°C for 48 hours for aerobic bacteria, and at 25°C  $\pm$  1°C for yeasts and fungi. Following incubation, enumeration of total colony-forming units (CFU) from each plate was performed by a third-party lab and colonies were evaluated for organism identification using standard techniques, but susceptibility testing was not performed. Proportion of cultures which showed no growth/sterile was also recorded. ATP readings were measured in terms of relative light units (RLU) (3M Clean-Trace, Maplewood, MN).

### *Statistical Analysis*

ATP bioluminescence values (RLU) levels and CFU counts were used to describe microbial load in the control and intervention arms. ATP levels were converted to log 10 to normalize distribution. To determine significance of differences in mean ATP levels, an independent *t* test was used for comparing groups, and ANOVA was used for comparing collection areas and sampling dates. ANCOVA was used for multivariate analysis, to include date and area of collection. To determine significance of differences in mean CFU counts, Poisson regression was used for both the univariate and multivariate comparisons. *P*-values less than .05 was considered significant. All statistical analysis was performed using SAS 9.4 (Cary, NC).

### *Safety Monitoring*

Throughout the duration of the study, all patients exposed to DHP were monitored by hospital staff for any adverse symptoms associated, including irritation of the eyes, skin, nose, or throat, difficulty breathing, headache, dizziness, loss of consciousness, or change in hair color.<sup>19</sup> Additionally, all patients (old enough to respond), parents, or guardians were interviewed and responses were recorded in patients' medical records regarding any adverse symptoms experienced.

## **Results**

We collected and analyzed a total of 280 surface cultures and ATP surface swabs. The overall mean surface microbial burden was significantly reduced in the intervention group when compared to the control group as seen in Figure 1 (mean 5.50 vs 11.77, *P*<.001).

Additionally, reductions in microbial CFUs were seen across all sampling sites in the intervention group as shown in Figure 2. ATP readings in both the control and intervention groups showed passing levels of surface cleanliness however, as seen in Figure 3, a reduction in the mean RLU levels was also noted in the intervention group when compared to the control group (172.08 vs 225.83, *P*<.006). A surface area is considered clean when ATP reading is below 250 RLU based on the manufacturer's instructions for use.<sup>23</sup>



## Safety Monitoring

Eighteen patients were exposed to DHP during the course of the study (Table 1). The patients' ages ranged from 16 months to 19 years old with a mean of 10.56 years. The 2 age categories that included the largest proportion of the patients were 7-9 years (27.8%) and 13-15 years old (27.8%). Regarding sex, 8 (44.4%) of the patients included in the study were male, and 10 (55.6%) were female. The mean length of stay for the patients included in the study was 4.83 days, with 12 (66.7%) of the patients staying 1-3 days, and the longest stay being 13 days. During the course of their stay, patients together with their parent(s) or guardian(s) were evaluated for any adverse symptoms associated with DHP exposure, including irritation of the eyes, skin, nose, or throat, difficulty breathing, headache, dizziness, loss of consciousness, or change in hair color.<sup>19</sup> Additionally, review of all 18 patient records revealed that none of the patients or their parents or guardians reported any adverse symptoms associated with DHP exposure.

Air sampling analysis revealed a reduction in aerobic CFU in the intervention group when compared to the control group, but the reduction did not achieve statistical significance ( $P = .139$ ).

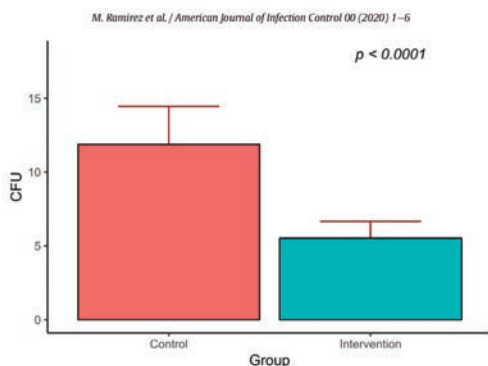


Fig 1. Mean surface CFU comparison by group.

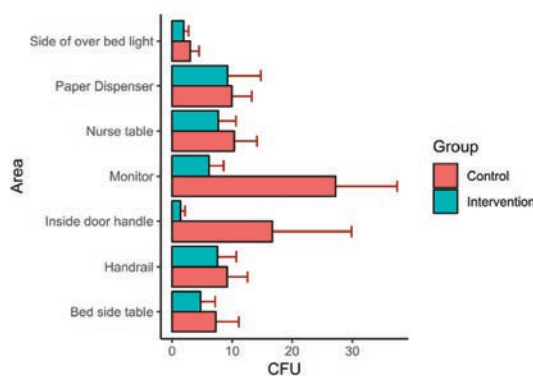


Fig 2. Multivariate Poisson regression analysis of CFUs by collection site and group.

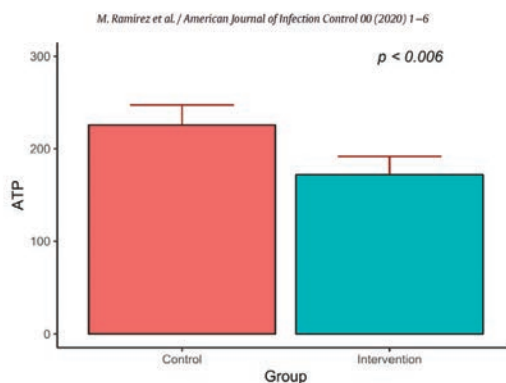


Fig 3. Mean surface ATP comparison by group.



## Discussion

The results of this study demonstrate that DHP was effective in reducing the residual microbial bioburden on surfaces and in the air, though reductions in the air did not reach statistical significance. We collected the air samples at a singular time point on a weekly basis during the course of the study. The results of the air samples could have been influenced by several factors, including the amount of traffic movement, temperature, humidity, and levels of bioload particles or organisms present in the space.<sup>24</sup>

Our findings are in contrast to an experimental study of DHP targeted against 3 multidrug-resistant organisms which found that there was no difference in reduction of the organisms tested between the control and DHP intervention group.<sup>25</sup> Our study was conducted in an active clinical setting and we sampled different surface areas over a period of several weeks. Furthermore, we did not alter any traffic or activities within the space or control extraneous environmental factors as outlined in the experimental study.

DHP is a novel delivery form of hydrogen peroxide in an occupied space. The use of hydrogen peroxide in health care has been well described as an effective disinfectant in its vapor form.<sup>3,17,26-28</sup> Hydrogen peroxide demonstrates potent antimicrobial activity against a broad spectrum of microorganisms, including those most commonly associated with health care-associated infections, spore-forming organisms, and mycobacteria.<sup>17,18,26-28</sup> This is achieved through microbial oxidation, leading to disruption of essential cell components, such as membrane lipids and DNA, and resulting in a loss of viability and infectivity.<sup>26,27</sup>

**Table 1**

Characteristics of subjects included in the prospective cohort study of eligible pediatric cancer patients at Unidad Nacional de Oncología Pediátrica (UNOP) in Guatemala City during the period of August 2, 2019-September 10, 2019

|                             |                        | All patients |      | Reported symptoms of overexposure to DHP <sup>a</sup> |   |
|-----------------------------|------------------------|--------------|------|-------------------------------------------------------|---|
|                             |                        | N            | %    | N                                                     | % |
| Age <sup>i</sup>            | 3 years old or younger | 2            | 11.1 | 0                                                     | 0 |
|                             | 4-6 years old          | 1            | 5.6  | 0                                                     | 0 |
|                             | 7-9 years old          | 5            | 27.8 | 0                                                     | 0 |
|                             | 10-12 years old        | 3            | 16.7 | 0                                                     | 0 |
|                             | 13-15 years old        | 5            | 27.8 | 0                                                     | 0 |
|                             | 16 years old or older  | 2            | 11.1 | 0                                                     | 0 |
|                             | Missing                | 0            |      |                                                       |   |
|                             | Mean age (y)           | 10.56        |      |                                                       |   |
| Sex                         | Male                   | 8            | 44.4 | 0                                                     | 0 |
|                             | Female                 | 10           | 55.6 | 0                                                     | 0 |
|                             | Missing                | 0            |      |                                                       |   |
| Length of stay <sup>j</sup> | 1-3 days               | 12           | 66.7 | 0                                                     | 0 |
|                             | 4-7 days               | 1            | 5.6  | 0                                                     | 0 |
|                             | 8-14 days              | 5            | 27.8 | 0                                                     | 0 |
|                             | Missing                | 0            |      |                                                       |   |

Note: Patient medical records are the source of all demographic data. Eligible patients included individuals who stayed overnight in the Intensive Care Unit at least one night during their stay in a room with the Synexis DHP unit installed and operating.

<sup>a</sup>Values tabulated from responses to a questionnaire given to patients and their parents upon discharge. Potential symptoms on the questionnaire included eye irritation, skin irritation, nose and throat irritation, breathing difficulty, headache, dizziness, loss of consciousness, and change in hair color.<sup>19</sup>

<sup>i</sup>Age was measured at discharge in years rounded down to the nearest whole number.

<sup>j</sup>Length of stay was measured in days, counting exclusively (ie, January 1-4 would be measured as 3 days).

The key difference between DHP and other forms of airborne hydrogen peroxide, such as hydrogen peroxide vapor (HPV) or mist, is its safety for use in occupied settings. DHP systems produce hydrogen peroxide at far more dilute concentrations than other airborne hydrogen peroxide systems, yet DHP demonstrates an effective microbiocidal activity because of its dry, nonaqueous gas state. Hydrogen peroxide and water are chemically very similar and are both attracted to the same sites on a microbe's surface. Thus, in the presence of water, hydrogen peroxide must be much more concentrated to outcompete water molecules in order to occupy those sites and effectively kill the microbe. While all forms of hydrogen peroxide compete with water on some level due to the polar nature of both molecules, the general absence of water in DHP allows it to be effective at low concentrations in comparison to aqueous hydrogen peroxide, which must be significantly more concentrated in order to have in impact in spite of the water in which it is



mixed. The presence of water is only a concern with DHP on a location by location basis (i.e., a small spill of water next to a sink), whereas it is always a concern for aqueous hydrogen peroxide because the water is an inherent component of the disinfectant. DHP systems achieve hydrogen peroxide concentrations well below OSHA's safety limit of 1.0 ppm<sup>19</sup> while HPV and mist systems produce concentrations as high as 338 and 160 ppm, respectively.<sup>29</sup> It is worth noting that the concentration of hydrogen peroxide achieved by DHP systems is also lower than the concentration of hydrogen peroxide naturally maintained by the lactoperoxidase cycle in the human respiratory tract.<sup>30-32</sup>

In this study, DHP achieved statistically significant microbial reductions in surface bioburden despite the fact that bioburden levels were relatively low to begin with. This was also reflected in ATP testing which has been shown to be an effective environmental cleaning assessment tool when total microbial counts are low.<sup>33</sup> Prior research has suggested that as few as 15 CFU may serve as an infectious threshold of contamination.<sup>33-37</sup> While, the mean microbial burden for 2 sampling sites—the monitor and inside of the door handle—exceeded this threshold in the control group, the mean microbial burden for all sites was well below this threshold in the intervention group. This not only demonstrates the efficacy of DHP, but aligns with the body of research demonstrating that manual cleaning as a stand-alone measure for mitigating environmental infection risk is not adequate.<sup>3</sup> Studies such as that by Shams et al in which *C. difficile* was recovered from 50% of patient rooms after manual cleaning or that by Chen et al which found that 55% of patient rooms remained contaminated with multidrug-resistant organisms, including MRSA and multidrug-resistant *Acinetobacter*, despite terminal manual cleaning and disinfection underscore this inadequacy.<sup>4,38</sup> Variability in both staff performance and compliance with cleaning protocols, material and product compatibility, and high environmental services staff turnover rates are among the many factors that may render manual cleaning efforts suboptimal. Automated, “no-touch” decontamination technologies have been developed to address this problem. Research has shown that 2 of the most commonly utilized of these technologies—ultraviolet-C light systems and HPV systems – can be a valuable adjunct to manual cleaning in achieving more thorough environmental decontamination, but they have several notable disadvantages.<sup>3,26</sup> First, neither system can be used in an occupied setting because of human exposure safety issues.<sup>3,26</sup> Thus, these technologies are commonly used for terminal disinfection.<sup>3,26</sup> These adjunct technologies while effective, only provides a transient level of disinfection for that particular time period. The reintroduction of a patient, patient care equipment and health care workers to that room inevitably leads to recontamination of that environment.<sup>3</sup> The cycle time for each system also varies and can present a challenge for patient throughput. Second, both technologies require substantial capital equipment costs in addition to staff overseeing its use. In the case of ultraviolet-C technology, studies have shown that its efficacy is dependent on a number of parameters that such as distance from the device, room size and shape, direct line of sight from the device, organic load, and dose delivered.<sup>26</sup> HPV systems on the other hand require the treated space to be sealed off in addition to being vacant, adding to the total cycle time.

DHP systems, by contrast, offer a continuous microbial reduction that can address in real-time the ongoing contamination of the health care environment without interrupting patient care. Once installed, it requires minimal staff oversight or operation and can be deployed anywhere in a facility. In this study, we used the stand-alone DHP units which were plugged in to a standard outlet and turned on. The DHP gas was generated, and diffused throughout the space, reaching and treating all areas within the room. Individual exposure to DHP showed no untoward side effects or adverse reactions during the course of the study (Table 1).



## Limitations

This study has some limitations. Identification of recovered organisms at the species level was not performed, which would help provide clinical relevance if the identified organisms were among those known to cause hospital-acquired infections. However, given that oncology patients are potentially susceptible to infection from microorganisms that are not pathogenic to the general population, a reduction in total bioburden is arguably clinically relevant in this population.<sup>39</sup> This study was also limited to 4 patient rooms. Future study utilizing a larger sample size over a longer period is needed in order to evaluate the clinical impact of DHP on the incidence of hospital-acquired infections.

## Conclusions

DHP was effective in reducing surface and air microbial bioburden in an occupied space. This adjunct environmental cleanliness and control strategy can be implemented without affecting patient throughput and staff workflow. Further studies on the impact of DHP decontamination on incidence of hospital-acquired infections should be performed.

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