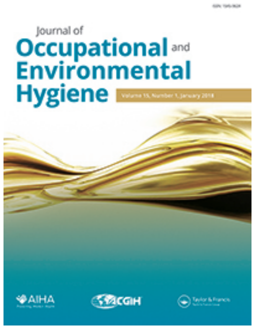

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# Ambulance disinfection using Ultraviolet Germicidal Irradiation (UVGI): Effects of fixture location and surface reflectivity

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**ABSTRACT**

Ambulances are frequently contaminated with infectious microorganisms shed by patients during transport that can be transferred to subsequent patients and emergency medical service workers. Manual decontamination is tedious and time-consuming, and persistent contamination is common even after cleaning. Ultraviolet germicidal irradiation (UVGI) has been proposed as a terminal disinfection method for ambulance patient compartments. However, no published studies have tested the use of UVGI in ambulances. The objectives of this study were to investigate the efficacy of a UVGI system in an ambulance patient compartment and to examine the impact of UVGI fixture position and the UV reflectivity of interior surfaces on the time required for disinfection. A UVGI fixture was placed in the front, middle, or back of an ambulance patient compartment, and the UV irradiance was measured at 49 locations. Aluminum sheets and UV-reflective paint were added to examine the effects of increasing surface reflectivity on disinfection time. Disinfection tests were conducted using *Bacillus subtilis* spores as a surrogate for pathogens.

Our results showed that the UV irradiance varied considerably depending upon the surface location. For example, with the UVGI fixture in the back position and without the addition of UV-reflective surfaces, the most irradiated location received a dose of UVGI sufficient for disinfection in 16 s, but the least irradiated location required 15 hr. Because the overall time required to disinfect all of the interior surfaces is determined by the time required to disinfect the surfaces receiving the lowest irradiation levels, the patient compartment disinfection times for different UVGI configurations ranged from 16.5 hr to 59 min depending upon the UVGI fixture position and the interior surface reflectivity. These results indicate that UVGI systems can reduce microbial surface contamination in ambulance compartments, but the systems must be rigorously validated before deployment. Optimizing the UVGI fixture position and increasing the UV reflectivity of the interior surfaces can substantially improve the performance of a UVGI system and reduce the time required for disinfection.

**KEYWORDS**
Ambulance; decontamination; emergency medical services; health care-associated infection; infection control



## Introduction

Ambulance patient compartments are frequently contaminated with pathogenic microorganisms shed by patients during transport. These microorganisms can potentially be transferred to subsequent patients and to emergency medical service (EMS) workers by direct contact with the surfaces or by indirect transmission via hands or medical items. The potential risk for the transmission of infections via contaminated surfaces is of great concern to the EMS community and could become a critical problem during an infectious disease pandemic, when large numbers of highly contagious patients would be transported and when the ability to decontaminate ambulances and return them to service as soon as possible would be needed. In 2012, the InterAgency Board, a working group of emergency preparedness and response officials, listed the development of rapid decontamination systems for ambulances as one of its research priorities.[1]

Current procedures for infection prevention in emergency medical services typically call for the interior of

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the patient compartment to be cleaned of visible bodily fluids or soil, sprayed with an approved disinfectant, wiped after the appropriate contact time, and allowed to air dry.[2] Such manual disinfection efforts are tedious and time-consuming, and several studies have shown that persistent contamination is common in patient compartments even after cleaning. Roline et al.[3] swabbed five potential fomite surfaces in ambulances and found methicillin-resistant *Staphylococcus aureus* (MRSA) in 10 of 21 ambulances that were examined. Brown et al.[4] sampled 16 areas within ambulances used in rural Maine and found MRSA in 25 of 51 ambulances studied. Rago et al.[5] found *S. aureus* in 69% (49/71) of ambulances serving a metropolitan area; 77% (77/100) of the isolates were resistant to at least one antibiotic, and 34% (34/100) were resistant to two or more antibiotics. Valdez et al.[6] seeded two high-contact surfaces with a bacteriophage and found that the contamination was spread to 56% (27/48) of other commonly touched surfaces that were examined, primarily by hand contact. Standard cleaning practices only partly reduced the contamination. A study of Ohio EMS personnel found that 4.6% (13/280) had nasal carriage of MRSA, suggesting that occupational exposures were resulting in colonization of workers by pathogens.[7]

One potential method to reduce the risk of disease transmission in patient compartments is disinfection through the use of whole-compartment systems, such as disinfectant foggers or ultraviolet germicidal irradiation (UVGI). Such germicidal systems are employed as a final disinfection step after the patient has been removed and visible contamination has been cleaned (called terminal disinfection). Terminal disinfection systems do not eliminate the need to first manually clean heavy or visible contamination. However, the use of a terminal disinfection system could allow the manual cleaning to focus on surfaces that are visibly contaminated or most prone to contamination, rather than attempting to wipe down every surface in the ambulance.

UVGI has been studied as a method for surface disinfection in hospital rooms (reviewed by Weber et al.[8]). Anderson et al.[9] cultured surface samples before and after UVGI treatment from 39 patient rooms that had been occupied by patients infected or colonized with vancomycin-resistant enterococci (VRE), *Clostridium difficile*, or *Acinetobacter*. They found that the use of a UVGI system reduced the numbers of viable pathogens at nine environmental sites by 97% ($p < 0.0001$). Jinadatha et al.[10] compared high-touch surfaces disinfected by manual cleaning to those disinfected by UVGI in 20 hospital rooms occupied for at least two days by patients with MRSA. Surfaces treated by UVGI had a significantly greater reduction in MRSA and other heterotrophic bacteria than did surfaces cleaned manually (adjusted incident rate ratio [IRR] = 7 for MRSA with a 95% confidence interval of <1–41; for HPC, IRR = 13; 95% CI = 4–48). Nerandzic et al.[11] showed that, on high touch surfaces, UVGI treatment in hospital rooms for 10 minutes reduced the number of colony-forming units (CFUs) of *Clostridium difficile* by 72%, MRSA by 99%, and VRE by 75% ($0.55 \pm 0.34$, $1.85 \pm 0.49$, and $0.6 \pm 0.25$ $\log_{10}$ CFU/cm$^2$, respectively). Pegues et al.[12] found that the use of a UVGI system substantially reduced the incidence of *C. difficile* infections in an adult hematology-oncology unit (IRR = 0.49; 95% CI = 0.26–0.94; p = 0.03). Rock et al.[13] showed that UVGI was highly effective at killing carbapenem-resistant enterobacteriaceae (CRE) inoculated onto high touch surfaces in an empty patient room ($10^6$ reduction in CFUs). In a recent large multi-hospital clinical trial, Anderson et al.[14] found that adding UV disinfection to the standard cleaning practices decreased the incidence of MRSA, VRE and multidrug-resistant *Acinetobacter* in exposed patients (IRR = 0.70, 95% CI = 0.50–0.98; p = 0.036).

UVGI systems have several potential advantages for terminal disinfection. They are relatively simple and easy to use, and do not leave chemical residues or risk exposing patients and workers to toxic chemicals. In ambulances, UVGI systems can be used while the crew cab is occupied, while fogging systems cannot. However, UVGI systems also have an important limitation: Because most materials are not good reflectors of ultraviolet light at germicidal wavelengths (primarily 254 nm), UVGI systems are less effective against microorganisms on surfaces that are not in a direct line-of-sight of the system.[15,16] UVGI and other terminal disinfection systems are also ineffective against microorganisms on surfaces that are covered or inaccessible, such as areas behind closed cabinet doors or retracted seat belts. One approach to improving the performance of these systems is to cover surfaces with UV-reflecting materials or coatings in order to better irradiate shadowed areas. Rutala et al.[17] painted the walls of a patient room in a hospital with UV-C reflective paint and found that the time required to decontaminate the room was reduced from 25 min to 5 min for *Staphylococcus aureus* and from 44 min to 9 min for *Clostridium difficile* spores. Jelden et al.[18] reported similar results.

Although terminal disinfection systems are increasingly popular for use in patient rooms in hospitals,[8] more information is needed about how well they work and how best to use them. The Centers for Disease Control and Prevention (CDC) recommends against the use of disinfectant fogging systems employing formaldehyde, phenol-based agents, or quaternary ammonium compounds in patient-care areas.[19] The CDC makes no recommendation regarding the use of systems based on hydrogen peroxide fogging, UVGI or ozone mists and says that more research is needed.[19,20] The Canadian Agency for Drugs and Technologies in Health (CADTH) also determined that insufficient evidence was available to make recommendations about terminal disinfection using vaporized hydrogen peroxide or UVGI.[21]

While information about the use of UVGI in hospital rooms is limited, information about the use of UVGI in ambulances is virtually non-existent. Although UVGI systems are being marketed as a means of surface disinfection in ambulances, a search of the biomedical literature located only one study which mentioned that UVGI was used for terminal disinfection of ambulances used for transportation of patients with Ebola virus disease, and in that case no test results or details about the system were provided.[22] Other than that work, no published peer-reviewed studies of UVGI in ambulances were found. Before UVGI systems can be reliably used in ambulances, much more information is needed about their efficacy and limitations in this application.

The purpose of this project was to test the ability of an ultraviolet germicidal irradiation system to disinfect the interior of an ambulance patient compartment, to examine the variations in irradiance among different locations in the compartment, and to study the effects of the location of the UVGI fixture and the addition of UV-reflective material or UV-reflective paint on the efficacy of the UVGI system. The information provided by this study will help to better understand the uses and limitations of UVGI systems in ambulances.

## Methods

### Ambulance

Our study was conducted using a 2005 Wheeled Coach Type III ambulance which met Federal Specification KKK-A-1822D when constructed.[23] For the purposes of evaluating surface disinfection, this specification is very similar to the construction standards for ambulances that are maintained by the National Fire Protection Association[24] and ASTM International.[25] A schematic of the ambulance patient compartment is shown in Figure 1. The patient cot was removed from the ambulance for all experiments.

### Ultraviolet germicidal light fixture

The ultraviolet germicidal light fixture used in these experiments was custom-built. It consisted of ten UV-C lamps with a primary wavelength of 254 nm (TUV PL-L 60 W/4P HO 1CT/25, Philips Lighting). Each lamp had a nominal wattage of 60 watts and a UV wattage of 12.4 watts. The lamps were mounted vertically in two circles (one upper, one lower) around an 11.1 cm (4 3/8″) diameter aluminum-covered post. The lamps were powered by five electronic ballasts (PureVOLT IUV-2S60-M4-LD, Philips Lighting). Photographs and a diagram of the fixture are included in the online supplemental material.

The light fixture was placed in three positions in the patient compartment for testing: Front, with the light 81 cm (32″) from the left interior wall and 217 cm (85 1/2″) forward of the rear doors of ambulance; Middle, with the light centered laterally between the interior walls and 132 cm (52″) from the rear doors; and Back, with the light position centered laterally between the interior walls and 48 cm (19″) from the rear doors. The light fixture positions are shown in the ambulance patient compartment schematic in the online supplemental information.

### Irradiance measurements

Irradiance measurements were made at 49 locations throughout the patient compartment as shown in Figure 1. A descriptive list of the locations is given in Table 1; photographs and an interactive schematic showing the sensor locations are included in the online supplementary material. The sensor locations were chosen to emphasize two areas: (1) surfaces that are frequently touched such as controls, handles, latches and hand rails and (2) surfaces that were not directly exposed to light from the UVGI fixture, such as between the primary care seat and the wall, and in the passageway leading to the front cab.

The irradiance measurements were made using 17 UV-C sensors (SED033-25, International Light Technologies) routed through three automated switch boxes (A803, International Light Technologies) connected to three radiometers (ILT-1700, International Light Technologies). A scan delay of 3 s was used for the switch boxes to prevent measurements from one sensor from being carried over to the next sensor reading.

To perform irradiance measurements, the sensors were placed at 17 locations. The UVGI light fixture was turned on and allowed to stabilize for 30 min. Separate irradiance measurements were collected with the fixture in the back, middle and front positions. The fixture was then turned off and 16 of the sensors were moved to different locations; one sensor was left in the same location (B2) for all tests to allow comparison of irradiance levels from different tests. The measurements were collected using the three light fixture positions. The fixture was turned off, 16 sensors were moved to the final set of locations and the measurements were repeated. The entire set of measurements was repeated three times with the sensors rotated among the locations, so that measurements were collected

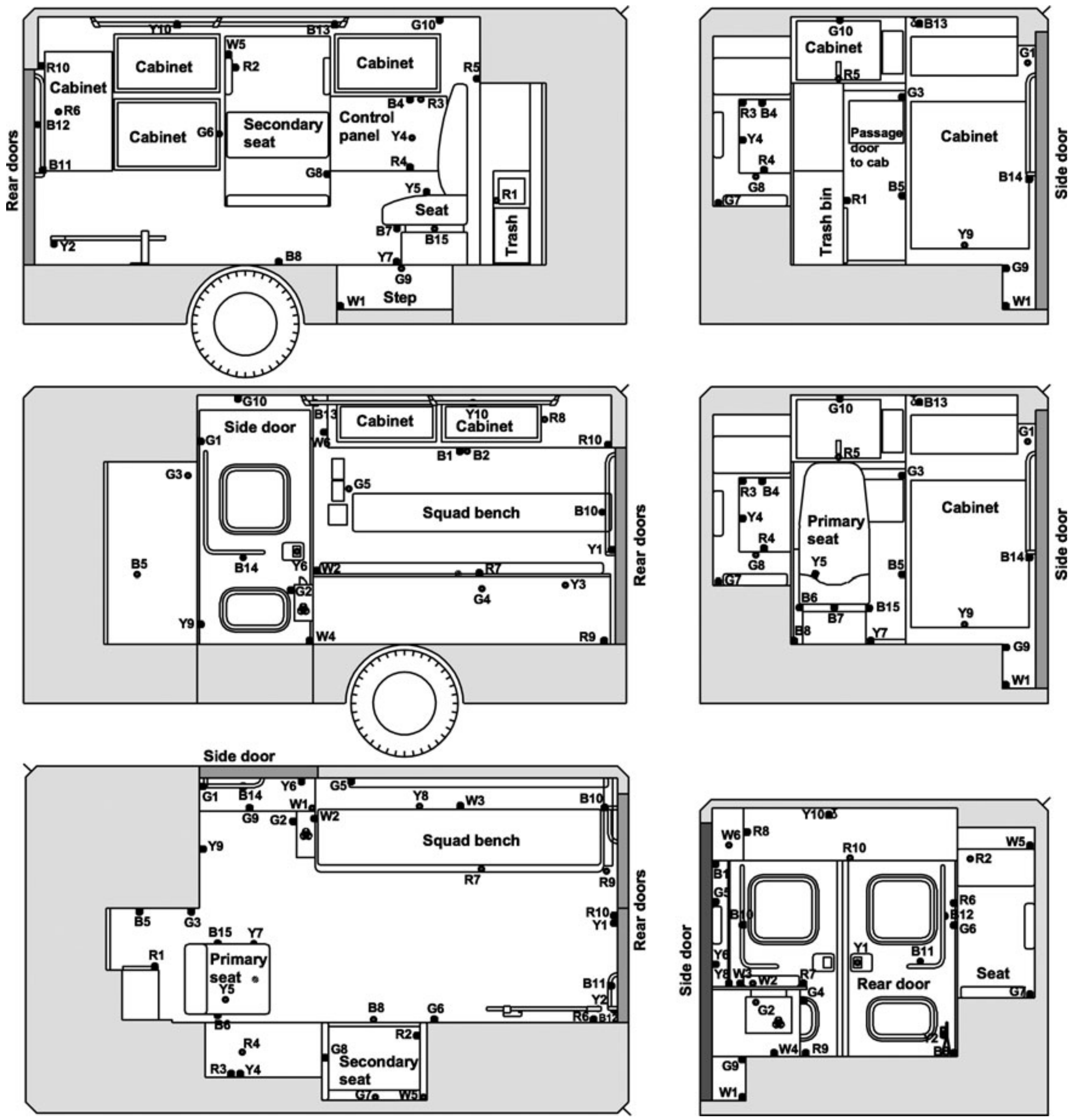


Figure 1. Ambulance patient compartment showing the locations of the UVGI sensors. A more detailed color schematic and photographs of the ambulance patient compartment are provided in the online supplemental material.



three times with different sensors at each location for each UVGI fixture position (153 data points).

To prevent exposure of personnel to UV-C, the exterior of the patient compartment windows was covered with aluminum foil, warning signage was placed on the ambulance doors and access to the ambulance was restricted during experiments. One person was allowed to enter the ambulance to move the UVGI fixture while it was on; this individual was completely covered in protective clothing and wore a UV-C absorbing face shield.

## *Ambulance interior surface reflectivity*

To examine the effects of surface reflectivity, three different ambulance interior surface conditions were tested. The first condition (called "Original") consisted of the ambulance interior surfaces as originally supplied by the ambulance manufacturer. The side panels and ceiling were primarily white melamine with aluminum trim and vinyl-covered padding on the edges. The cabinet sliding doors were clear plastic, the access doors were largely covered in diamond-plate aluminum, and the floor was a dark grey non-skid plastic material. Seat cushions were covered with blue vinyl.

For the second interior surface condition (called "Reflective"), many of the vertical white melamine surfaces were covered in diamond-plate aluminum sheets to increase the surface reflectivity for UV light within the patient compartment and thereby increase the irradiation of shadowed areas. Diamond-plate aluminum sheets also

**Table 1.** Descriptive list of UVGI sensor locations in the ambulance patient compartment. The locations are shown schematically in Figure 1 and in the online supplemental materials.

| *Sensor ID* | *Sensor location* |
|---|---|
| B1 | Under center of high white cabinet on right side of ambulance, facing down |
| B2 | Above center of back cushion on squad bench seat, facing out |
| B4 | On light switch under cabinet by rear-facing primary patient care seat, facing down |
| B5 | On right wall of passageway to cab, facing left, straight across from R1 |
| B6 | Under right side of rear-facing primary care seat between seat and wall, facing wall |
| B7 | Under front edge of rear-facing primary patient care seat, facing floor |
| B8 | On floor centered in front of secondary patient care seat, facing ceiling |
| B10 | Vertical hand rail on right rear door, facing right wall |
| B11 | Horizontal hand rail on left rear door, facing door |
| B12 | Vertical hand rail on left rear door, facing door |
| B13 | Front of overhead hand rail, facing ceiling |
| B14 | Horizontal hand rail of side door, facing door |
| B15 | Underneath left side of rear-facing primary patient care seat, facing side door |
| G1 | On forward wall next to side door, facing back |
| G2 | On biohazardous sharps disposal box, facing forward |
| G3 | High on right side of passageway to cab, facing left |
| G4 | Next to middle seat belt buckle on front of right-wide squad bench, facing left |
| G5 | Next to oxygen connections on right wall beside side door, facing left |
| G6 | Left wall beside cabinet doors, facing right |
| G7 | Behind horizontal cushion on secondary patient care seat, facing up |
| G8 | On left side of secondary patient care seat alcove, facing back |
| G9 | On side door step, facing door |
| G10 | On ceiling above rear-facing primary patient care seat, facing floor |
| R1 | Next to door of trash bin in passageway to cab, facing right |
| R2 | On upper right cushion next to secondary patient care seat, facing forward |
| R3 | On top of control panel next to rear-facing primary patient care seat, facing right |
| R4 | On work counter in front of control panel next to rear-facing primary care seat, facing up |
| R5 | On latch of storage bin above rear-facing primary patient care seat, facing back |
| R6 | On latch of rearmost cabinet on left side, facing right |
| R7 | In front of cushion for squad bench, facing up |
| R8 | Next to rear-most upper cabinet on right side, facing left |
| R9 | On floor in front of right-side squad bench seat, next to rear door, facing up |
| R10 | On wall above center of rear doors, facing front |
| W1 | On bottom step at side door, back left corner, facing up |
| W2 | On squad bench above biohazardous sharps disposal box, facing forward |
| W3 | Center of back surface of right squad bench cushion, facing right wall |
| W4 | On floor below sharps disposal box, 7.5″ from right edge of squad bench, facing up |
| W5 | On top of left vertical cushion above secondary patient care seat, back corner, facing up |
| W6 | Center of end of right upper cabinet, facing forward |
| Y1 | On latch of left rear door, facing forward |
| Y2 | On handle of patient cot floor lock, facing floor |
| Y3 | Next to rear-most seat belt buckle on front of right-side squad bench, facing left |
| Y4 | Above control switches on control panel next to rear-facing primary care seat, facing right |
| Y5 | On horizontal cushion of rear-facing primary patient care seat, facing up |
| Y6 | On latch of side door, facing left |
| Y7 | Under right corner of rear-facing primary patient care seat, facing ceiling |
| Y8 | Behind cushion for left-side secondary patient care seat, facing up |
| Y9 | Next to latch for roll-up door on front right storage cabinet, facing back |
| Y10 | On back of overhead hand rail, facing ceiling |

were added to the floor beneath the rear-facing primary seat and the patient cot release handle. The positions of the added reflective material are shown in the online supplemental material.

For the third interior surface condition (called "UV paint"), the added aluminum sheets on the walls were removed, while the original aluminum surfaces and the aluminum added beneath the rear-facing primary seat and the patient cot release handle were left in place. All of the white melamine interior surfaces, including the walls, ceiling, and counters, were then painted with UV-C reflecting paint (UVC-MAX, product # W15984, Lumacept, Inc.). The floor, the original aluminum sheets and trim, and the seats and padding were not painted and remained in their original state.

### *Tests of disinfectant effects of UVGI*

The antimicrobial capabilities of the UVGI system were tested using *Bacillus subtilis* spores as a surrogate for pathogenic microorganisms. *B. subtilis* spores are recommended for use in the testing of sterilizing agents by the US Environmental Protection Agency (EPA) and the European Committee for Standardization (CEN).[26,27] *B. subtilis* spores also tolerate drying, washing and other experimental stresses well, which helps provide consistent experimental results. Since it is not a human pathogen, *B. subtilis* can be handled using Biosafety Level 1 precautions.

The EPA defines a disinfectant as "a substance, or mixture of substances that destroys or irreversibly inactivates bacteria, fungi and viruses, but not necessarily bacterial spores, in the inanimate environment."[28] Published results for a variety of pathogenic vegetative bacteria and viruses indicate that a UVGI dose sufficient to inactivate 99.9% of *B. subtilis* spores on a solid surface would also inactivate at least 99.999% of many pathogens, including *Staphylococcus aureus, Pseudomonas aeruginosa, Salmonella enteritidis, Serratia marcescens, Mycobacterium tuberculosis*, and human cytomegalovirus (although it should be noted that fungi are reported to be more resistant to UVGI).[15] Thus, for our experiments, we defined the surface disinfection dose as the UV-C dose sufficient to inactivate 99.9% of *B. subtilis* spores.

*Bacillus subtilis subsp. spizizenii* were purchased (Catalog #6633, American Type Culture Collection) and spores were produced following the procedure described in European Standard EN 14347:2005.[27] A detailed protocol for spore production is provided in the online supplemental material.

To determine the dose-response relationship between UV-C and *B. subtilis* spore inactivation, spores were exposed to UV-C using a system described previously.[29] The system was modified by adding a shutter that covered the UV-C lamp until its output stabilized. For exposure tests, 100 µL droplets of water containing 3–4 × $10^7$ spores were placed on 2.5 cm × 1.9 cm coupons made of type 316 stainless steel and allowed to dry at room temperature. The coupons were then exposed in sets of 3 to UV-C doses of 8, 16, 32, 64, or 128 mJ/$cm^2$. The experiment was repeated three times for a total of nine coupons exposed to each UVGI dose.

After exposure, each coupon was placed in a 6-well culture plate. Five ml of sterile 0.1% Tween 20 solution (Fisher Scientific) was placed in each well and the plates were shaken for 15 min using a titer-plate shaker (Model 4625, Thermo Scientific) at intensity level 5. 20 µL of each control sample and 100 µL of each UV-exposed sample were then plated onto tryptic soy agar plates (236950, Becton Dickinson) in an exponential spiral using an automated plater (Autoplate, Spiral Biotech). The plates were incubated overnight at 30°C and colony-forming units were enumerated using an automated plate counter (Qcount Model 510, Spiral Biotech).

The UVGI irradiance (intensity) and spore inactivation data were fitted to an exponential decay model of the form[15,30]

$$S = e^{-kIt}$$

where
S = fraction of spores surviving after UV-C exposure
k = rate constant ($cm^2$/mJ)
I = UV-C irradiance (mW/$cm^2$)
t = exposure time (seconds)

The UVGI dose in mJ/$cm^2$ was determined by integrating the irradiance over the exposure time. The UVGI dose required to disinfect a surface was calculated based upon the k-value found in the laboratory exposure experiments. This surface disinfection dose was divided by the irradiance measurements in the ambulance to calculate the exposure time required to disinfect each location in the compartment (the disinfection time) with the different fixture positions and surface reflectivities. The disinfection dose was used to convert irradiance levels to disinfection times when presenting the results because the disinfection time is easier for the reader to interpret. Irradiance levels for all experiments are provided in the online supplemental materials.

To test the ability of the UVGI system to inactivate microorganisms in the ambulance, spore-loaded coupons were fastened next to the inlet domes of the ultraviolet light sensors. Ten coupons were placed in the locations in the ambulance with the lowest irradiance levels for each interior surface type. Two control coupons were not exposed to UVGI but were otherwise treated in the same manner. The test was performed 6 times with the original interior surfaces, three times with the reflective interior surfaces, and three times with the UV-reflective paint on the interior surfaces. For all of these experiments, the UVGI fixture was placed in the back position for first half of the exposure time and in the front position for the second half.

## Results

### UV-C dose and Bacillus subtilis inactivation

The dose-response curve for *Bacillus subtilis* spores dried onto stainless steel coupons resulted in a rate constant (k) of 0.131 $cm^2$/mJ for the best-fit exponential decay curve with a goodness-of-fit of $R^2 = 0.930$. The average temperature in the exposure box during these experiments was 22.2°C (SD 0.3) and the average relative humidity was 57.0% (SD 2.5%). Based on these results, a UV-C dose of 52.6 mJ/$cm^2$ was required to inactivate 99.9% of the spores on a coupon (0.1% survival). This disinfection dose was used with the UV-C irradiance levels measured with the radiometers to calculate the disinfection times in the results presented below.

### UV disinfection time at different locations within the ambulance patient compartment

The shortest, median, and longest exposure times required for disinfection among the different locations in the ambulance are shown in Table 2 for each UVGI fixture position and interior surface type. Locations that

**Table 2.** Shortest, median, and longest disinfection times among the 49 test locations for each interior surface and UVGI fixture position. Note that the shortest time is given in seconds, while the other times are given in minutes. The longest disinfection time is the time required to inactivate 99.9% of the *B. subtilis* spores at the location with the lowest irradiation level, and thus is the estimated time required to disinfect all surfaces in the ambulance patient compartment.

| *UVGI fixture position* | *Interior surfaces* | *Shortest disinfection time (s)* | *Median disinfection time (min)* | *Longest disinfection time (min)* | *Location corresponding to longest disinfection time* |
|---|---|---|---|---|---|
| Front | Original | 44 | 6.7 | 990 | B6 |
| | Aluminum sheets | 42 | 6.3 | 156 | B6 |
| | UV-reflective paint | 32 | 3.6 | 117 | B6 |
| Middle | Original | 20 | 7.0 | 216 | B6 |
| | Aluminum sheets | 18 | 5.7 | 179 | W1 |
| | UV-reflective paint | 15 | 3.9 | 79 | W1 |
| Back | Original | 16 | 15.6 | 900 | W5 |
| | Aluminum sheets | 17 | 12.1 | 963 | W5 |
| | UV-reflective paint | 14 | 6.8 | 181 | G9 |
| 1/2 Front | Original | 29 | 5.0 | 244 | B6 |
| 1/2 Back | Aluminum sheets | 32 | 4.8 | 81 | B13 |
| | UV-reflective paint | 26 | 3.2 | 67 | B6 |
| 1/3 Front | Original | 37 | 6.9 | 234 | B6 |
| 1/3 Middle | Aluminum sheets | 39 | 5.0 | 79 | W5 |
| 1/3 Back | UV-reflective paint | 32 | 3.0 | 59 | B6 |

were further away from the UVGI fixture or in shadowed areas received much lower UV-C doses than did locations closer to the UV fixture and directly in the line-of-sight. For this reason, the longest disinfection times were 220–3400 times larger than the shortest times (Table 2). As a practical matter, the longest disinfection time determines the estimated overall time required to disinfect all locations in the interior of the ambulance. The overall disinfection time ranged from 16.5 hr for the poorest performing configuration to 59 minutes for the best.

Figure 2 shows the disinfection times at each of the 49 test locations in the ambulance with the UVGI light in the middle position and with the original interior surfaces. In this experiment, the disinfection times ranged from 20 s at location G6 (on the left wall directly facing the UVGI fixture) to 216 min at location B6 (between the rear-facing primary patient care seat and the left wall, facing the wall), indicating that location G6 receives 652 times as much UV-C light as does location B6 with this configuration.

### *UV disinfection time with different fixture positions*

The disinfection times when the UVGI fixture was in different positions in the ambulance and with the original interior surfaces are shown in Figure 3. The locations presented in the figure include the five worst locations (that is, the locations with the longest disinfection times) for each light position. Note that, as the light is moved to different positions, the disinfection times for some locations improve while others become worse. For example, when the light was moved from the front to the back, the disinfection time for location B12 (vertical hand rail on left rear door) decreased from 72 min to 7 min, while for location W1 (bottom step at side door) the disinfection time

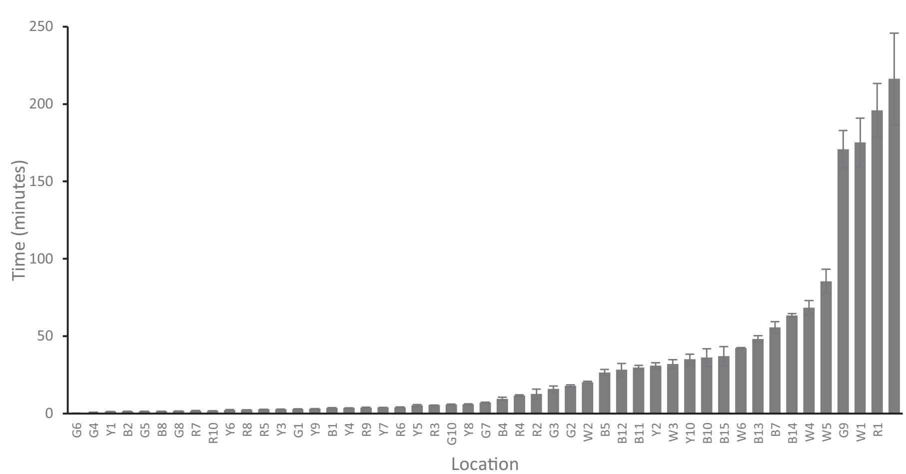


**Figure 2.** Surface disinfection time (exposure time required to inactivate 99.9% of *Bacillus subtilis* spores) at each location in the ambulance with the UVGI fixture in the middle position and with the interior surfaces in their original condition (no aluminum sheets or UV-reflective paint added). Each bar shows the average of three experiments. Error bars show the standard deviation.

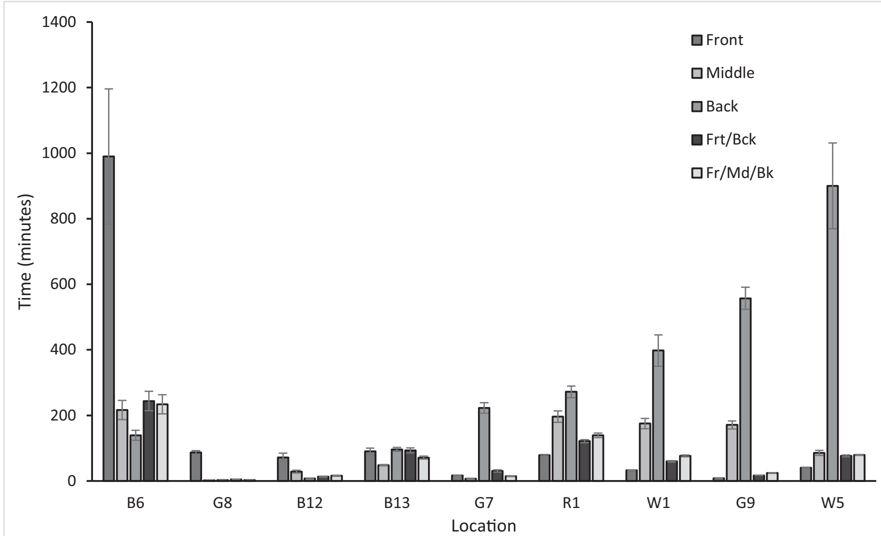


**Figure 3.** Effect of UVGI light position on the surface disinfection time at different locations. These experiments were conducted with the original interior surfaces and the UVGI fixture in the front, middle, or back position. The "Frt/Bck" data was calculated assuming that the fixture was in the front position for half of the exposure time and was moved to the back position for the other half. The "Fr/Md/Bk" data was calculated assuming that the fixture was in the front, middle and back positions for 1/3 of the exposure time each. The locations shown were chosen based on the 5 longest disinfection times for each light position. Each bar shows the average of three experiments. Error bars show the standard deviation.

increased from 32 min to 398 min. For the original interior surface condition, the worst overall disinfection time of 990 min occurred when the UVGI fixture was in the front position in the ambulance, and the best overall disinfection time of 216 min occurred when the fixture was in the middle position. The results for all interior surface conditions are summarized in Table 2.

### *Effects of the addition of UV-C reflective aluminum sheets or UV-C reflective paint*

The effects of adding aluminum sheets or UV-reflective paint to the interior surfaces are shown in Figure 4. The results presented in this figure were calculated assuming that the UVGI fixture was placed in the front, middle, and back positions for one-third of the exposure time each.

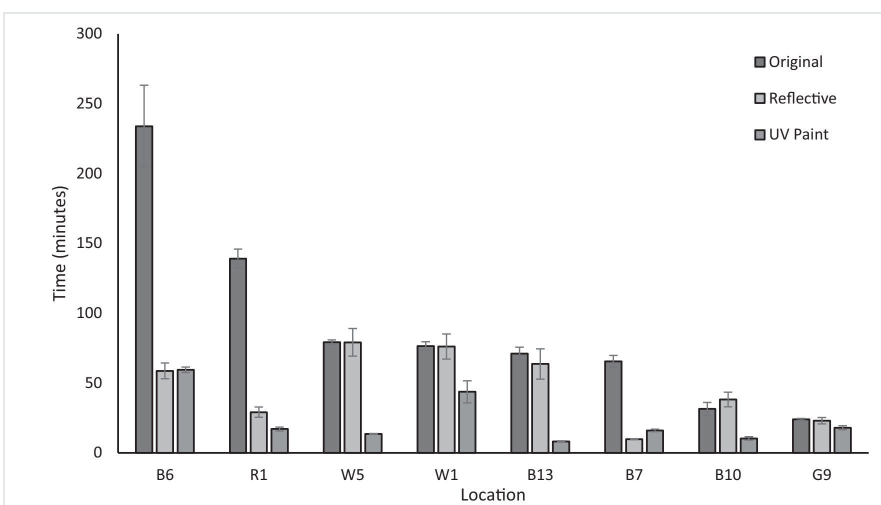


**Figure 4.** Effect of ambulance interior surface treatments on the surface disinfection time. These results were calculated assuming that the UVGI fixture was placed in the front, middle and back positions for one-third of the exposure time each. As described in the materials and methods section, original refers to the ambulance interior surfaces as they were originally made by the manufacturer. Reflective refers to the addition of reflective aluminum plating to various interior surfaces as shown in the online supplemental material. UV paint refers to coating the white melamine interior surfaces of the ambulance with white UV-C reflective paint. The locations shown were chosen based on the five longest disinfection times for each interior surface. Each bar shows the average of three experiments. Error bars show the standard deviation.

With the original interior surfaces, the time needed to disinfect all locations was 234 min. The overall disinfection time was reduced to 79 min by adding the reflective aluminum surfaces, and to 59 min by painting the interior with the UV-reflective paint.

### *Ambulance tests of UVGI system using Bacillus subtilis spores*

The survival fraction vs. UVGI dose for *Bacillus subtilis* spores on coupons placed in the ambulance is shown in Figure 5, along with the predicted survival curve from the laboratory experiments. The correlation coefficient between the laboratory model and the ambulance results was 0.804. After 20 min, the survival fraction of the spores in the ambulance ranged from 0.1–17.9% with the original interior surfaces and from below detection to 1.9% with the UV-reflective paint. The average temperature during these experiments was 21.1°C (SD 4.6) and the average relative humidity was 43.9% (SD 8.4%).

## Discussion

Ultraviolet germicidal irradiation is being marketed as a terminal disinfection method for ambulance patient compartments. Commercial portable UVGI systems range in price from around $600–3,800, depending upon the size and power of the fixture and features such as timers, dose measurement systems and remote controls. UVGI is very effective at inactivating microorganisms if the microbes receive a large enough dose of ultraviolet light. However, ensuring that all surfaces within the compartment are sufficiently irradiated to disinfect them requires careful planning and testing and an appreciation of the limitations of UVGI systems.

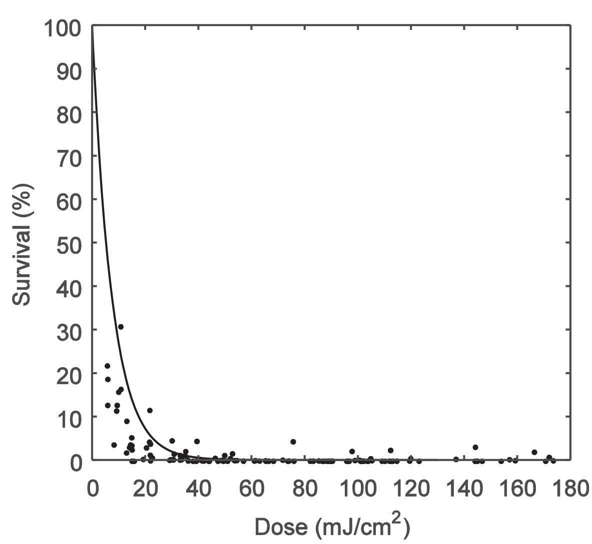


**Figure 5.** Survival of *Bacillus subtilis* spores exposed to UVGI in the ambulance interior. Each coupon of spores was mounted alongside the sensor dome of a radiometer, which was used to measure the UVGI dose received by the coupon. The solid line is the predicted survival based on the laboratory tests. The dots show the results from 120 coupon tests in the ambulance.

### *Surface location and disinfection time*

UVGI irradiation levels varied greatly from place to place within the ambulance compartment. As seen in Figure 2, this caused the disinfection times to vary considerably with surface location as well. Since the purpose of the UVGI system is to disinfect as much of the patient compartment as is practically possible, the time required for overall disinfection is driven by the locations with the lowest irradiance levels, not by the average irradiance. For this reason, interventions that increase the irradiance of the worst locations will reduce the overall disinfection times for the ambulance compartment and allow for a more rapid turnaround of the ambulance.

For each combination of UVGI fixture position and surface reflectivity, the median disinfection time among all locations was much less than the longest time (Table 2), indicating that the distribution of the disinfection times among the various locations was skewed. This can be seen in Figure 2; half of the locations had disinfection times of 7 min or less, and 90% had disinfection times of 69 min or less, but the exposure time needed to disinfect the worst location was 216 min. These results show that, when testing a UVGI system, UVGI measurements must be made at multiple locations. In addition, a particular effort should be made to find the locations with the lowest irradiance levels, since these locations will determine the exposure time needed to disinfect the entire compartment. On the other hand, this also suggests that the overall disinfection time can be lowered substantially by increasing the irradiance of a small number of areas that are most shadowed from the UVGI fixture, or by determining that some areas either do not require disinfection or that those areas will need to be disinfected by other means.

### *Effects of UVGI fixture position on disinfection time*

The position of the UVGI fixture had a substantial effect on the overall disinfection time for the compartment. With the original interior surfaces, moving the fixture from the front to the middle position reduced the disinfection time from 16.5 hr to 3.6 hr. The ratio of the longest time to the median time was also reduced from 148 to 31, suggesting that the light was more evenly distributed. Similar results were seen with the reflective material and the UV-reflective paint. Thus, determining the optimal location for the UVGI fixture is important when designing a UVGI system for an ambulance.

An additional step that can be taken to better distribute the UV light within the compartment is to move the UVGI fixture during disinfection. We examined two possibilities: (1) placing the fixture in the front position for half of the disinfection time and the back position for the other half; and (2) placing the UVGI fixture in the front, middle and back position for one-third of the disinfection time each. With original interior surfaces, moving the fixture did not improve the disinfection time compared to leaving the fixture in the middle position. However, with the reflective aluminum, the disinfection time was reduced from 156 min with the light in the front position to 81 min by splitting the exposure time between the front and back positions. With the UV-reflective paint, splitting the exposure time between the front and the back reduced the disinfection time from 79 min to 67 min compared to leaving the fixture in the middle, and splitting the time between the front, middle and back reduced the time to 59 min. Further reductions in the disinfection time could be achieved by using a more powerful UVGI fixture or using multiple fixtures for disinfection; using two fixtures, for example, would potentially cut the overall disinfection time in half.

### *Effects of different surface reflectivities on disinfection time*

One way to enhance the efficacy of UVGI fixtures is to increase the UV reflectivity of surfaces in the compartment so that more UV irradiation reaches locations that are not directly exposed. Most common materials reflect only a small amount of the UV-C. For example, ordinary white water-based paint is reported to reflect about 10–23% of UV-C light, while stainless steel reflects about 28% and glass only reflects about 4%.[15] By comparison, bare aluminum has a surface reflectivity of about 74–84% depending upon the finish, and the UV-C reflective paint that we used has a reflectivity of about 65%. Increasing the UV surface reflectivity with reflective aluminum sheeting or UV-C reflective paint both substantially reduced the disinfection times. As seen in Table 2, the best overall disinfection time with the original surfaces was 216 min. This was reduced to 79 min with the reflective aluminum and 59 min with the UV-reflective paint.

The UV-reflective paint reduced the disinfection times for all individual locations with all fixture positions, and gave the lowest overall disinfection times. The use of reflective surfaces was not quite as effective, probably because more surfaces were covered with the UV-reflective paint than with the aluminum sheeting, most notably the ceiling. The UV-reflective paint also provides a diffuse reflection, which may help distribute the UV light better, while the reflection from the aluminum sheets is specular (mirror-like). More surfaces could have been covered with the aluminum sheets, but the aluminum sheets have the disadvantage of creating visible light reflections and glare in the compartment. This might be reduced by using aluminum with a brushed or matte finish, but probably can't be eliminated entirely. On the other hand, the UV-reflective paint that we used is intended for hospital rooms and will not tolerate the repeated scrubbings that need to be performed on some surfaces in the ambulance compartment. Thus, a combination of aluminum sheeting and UV-reflective paint might provide the best method to optimize the performance of a UVGI system.

### *Limitations*

Our study has several limitations. First, and most important, it is not feasible to perform UV irradiance measurements at every possible location in the ambulance. We attempted to find and measure the locations that received the lowest amounts of UV irradiation, but it is entirely possible that some unmeasured places received less. It is also important to note that covered locations, such as underneath seat cushions or behind cabinet doors, receive no UV exposure at all.

Second, the disinfection time in our article is based on the use of *Bacillus subtilis* spores as a surrogate for pathogenic microorganisms. Although bacterial spores are generally considered to be hardier when exposed to UV light than vegetative bacteria or viruses,[15] data on the susceptibility of surface pathogens to UVGI are limited, and some pathogens may survive higher doses of UV-C light than *Bacillus subtilis* spores. Fungi also are reported to have a much higher tolerance for UV exposure than bacteria.[15]

Third, the aluminum sheets and UV-reflective paint were freshly applied before our experiments. In a real-world application, these materials would likely lose some reflectivity over time due to dirt, corrosion, abrasion, and other factors.

Fourth, we did not examine the effects of soiling such as by blood droplets or fecal matter on the efficacy of the UVGI system. Such opaque materials are known to block UV-C, and thus it is important that the ambulance be cleaned of any visible contamination before using UVGI as a final step in the disinfection process. Similarly, materials such as patient cots, sheets, papers, clipboards, clothing and other items should be removed from the patient compartment before beginning a UV disinfection cycle, since they will also shield surfaces from UV light.

Finally, although our general conclusions should be widely applicable, the quantitative results in our study are specific to the ambulance compartment that we tested. The irradiance levels, disinfection times, worst locations and other results that we found could be substantially different in ambulances with different UVGI fixtures, cabinet and seat arrangements, surface materials, etc. Thus, any ambulance UVGI system needs to be validated with the actual ambulance configuration for which it is to be used.

## Conclusions

Ultraviolet germicidal irradiation systems are potentially a useful tool for reducing the likelihood of infectious disease transmission in ambulance patient compartments. However, when implementing a UVGI terminal disinfection system in ambulances, the following points must be kept in mind.

- The amount of UV irradiation delivered to different surface locations can be expected to vary tremendously. When evaluating a UVGI system, multiple locations should be tested.
- The time required to disinfect an ambulance compartment is governed by the exposure time needed for the least-irradiated surfaces.
- Covered and concealed locations, such as underneath seat cushions or behind cabinet doors, will not be disinfected by a UVGI system.
- The position of the UVGI fixture can have a substantial effect on the disinfection time.
- Moving the UVGI fixture to multiple locations during a disinfection cycle or using multiple fixtures can reduce the disinfection time.
- Increasing the UV reflectivity of interior surfaces can also reduce the disinfection time.
- Before putting it into service, any ambulance UVGI system should be thoroughly tested with the actual ambulance configuration for which it is to be used, and should be periodically re-tested to verify that the UVGI system's performance has not changed over time.

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