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REVIEW

The microbiological impact of pulsed xenon ultraviolet disinfection on resistant bacteria, bacterial spore and fungi and viruses

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Pathogens can persist in the patient care environment and cause the risk of transmission to patients. Pulsed xenon ultraviolet (PX-UV) is increasingly being used to disinfect patient rooms, operating rooms and other areas. Data on the impact of PX-UV on resistant bacteria, bacterial spores, fungi, viruses and the Ebola virus are lacking in the literature. Laboratory data are presented in this paper on the log reductions observed after the exposure of PX-UV to a variety of organisms in a laboratory setting.

Keywords: hospital-acquired infections, HAIs, organisms, pathogens, pulsed xenon ultraviolet, PX-UV

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at ambient temperatures under the sterile laminar flow of the biosafety cabinet.

Two control Petri dishes and two Petri dishes treated in identical conditions were prepared for each organism, and placed on a mount at an angle of 45 degrees from horizontal, at approximately one metre in height, and at a distance of one metre from the PX-UV device. The exposure time was five minutes. The control Petri dishes remained covered and the treated ones remained open during exposure. A curve of exposure times was carried out at 0, 5, 10, 15 and 30 minutes for *A. niger*. Each exposure time was performed in triplicate.

After exposure, the control and treated Petri dishes were incubated at 37 °C for 24–72 hours, depending on the degree of growth of the colonies. The results were obtained by comparing the number of colonies in the treated and control samples.

The protocol was repeated on three different days, on three different degrees of culture growth, i.e. in biological triplicate. Therefore, there were six final figures for the treated and control samples.

Gram-positive bacterial spores

The following Gram-positive bacterial spores were used from commercially available sources:

- *Geobacillus stearothermophilus* ProSpore® spore kit (10^6 spores), in 4 ml ampoules with culture medium and growth indicator.
- *B. atrophaeus* spore kit (10^7/ml), in a 10 ml vial, with culture medium and growth indicator.

Middle East respiratory syndrome coronavirus

Viral suspensions, with an approximate titre of between 10^5 and 10^6 CFU/ml, were prepared in a Dulbecco's modified eagle medium (DMEM) culture, supplemented with 5% foetal bovine serum (FBS). The viral suspensions were applied in the form of a small drop of 500 µi in the centre of p-100 Petri dishes which had previously been prepared by applying a sheet of Parafilm® plastic film in order to reduce the surface tension of the drop so that it remained more exposed. Further optimisation was created in a follow-up experiment by replacing the 500 µi drop with 20 drops of 5 µi per Petri dish in order to reduce the shielding from the UV light produced by the liquid in the sample.

The exposure conditions were similar to those already described for the bacteria, with the exception that the liquid suspensions were placed directly on the working surface of the biosafety cabinet. A pair of control Petri dishes and a pair of treated Petri dishes were prepared under identical conditions for each exposure. After carrying out exposure to UV light, titration of the control and treated viral suspensions was performed, and their infectivity assessed in susceptible Vero E6 (African green monkey) cells through tests for the formation of lysis plaques. The viral titres in the samples were treated for differing periods and to determine the reduction in viral viability that exposure to PX-UV produced in Middle East respiratory syndrome coronavirus (MERS-CoV). The aforementioned protocol was repeated on the basis of three different prior growths of cultures, i.e. in biological triplicate.

Table 1: Independent laboratory testing results, involving pulsed xenon ultraviolet

Organism	Cycle time (minutes)	Distance (metres)	Pathogen count before disinfection	Pathogen count after disinfection	Logarithmic reduction measured
Klebsiella pneumoniae	5	1	1.88E+10	3.42E+01	8.74
Pseudomonas aeruginosa	5	1	9.12E+10	4.30E+01	9.33
Acinetobacter baumannii	5	1	6.07E+10	4.67E+01	9.11
Escherichia coli	5	1	3.32E+10	2.68E+01	9.09
Staphylococcus aureus	5	1	4.52E+10	3.47E+01	9.11
Geobacillus stearothermophilus	5	1	1.69E+06	2.57E+02	3.82
Bacillus atrophaeus	5	1	4.89E+05	2.51E+02	3.29
Aspergillus niger	5	1	1.07E+03	5.02E+02	0.33
Aspergillus niger	10	1	1.07E+03	1.37E+02	0.89
Aspergillus niger	15	1	1.07E+03	6.03E+01	1.25
Aspergillus niger	30	1	1.07E+03	4.10E+01	1.61
MERS-CoV (liquid)	5	1	4.13E+04	2.17E+04	1.54
Vaccinia virus (liquid)	5	1	4.98E+06	1.63E+05	1.38
IBDV (liquid)	5	1	2.41E+07	3.33E+06	0.86
VSV (dried)	5	1	2.60E+05	0.00E+00	All
Bacillus anthracis	15	1	4.5E+03	0.00E+00	All
Ebola virus	1	1	1.85E+07	0.00E+00	All

IBDV: infectious bursal disease virus, MERS-CoV: Middle East respiratory syndrome coronavirus, VSV: vesicular stomatitis virus

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the PX-UV device (8.7–9.1 log reduction) exceeds that which is necessary in the hospital environment. While the disinfection achieved was lower for the spore-forming organisms (3.29–3.82), it still exceeded the likely level of contamination in a hospital and patient care environment.

Additional time was required for disinfection of the fungus, *A. niger*, in order to reach a level of 1.61 CFU/ml log reduction at 30 minutes. *A. niger* is more resistant to UV light than vegetative organisms as it is a multicellular eukaryotic microbe, so these results were not unexpected.

Logarithmic reductions achieved for MERS-CoV, the vaccinia vaccinia virus and IBDV, were 0.58, 1.66 and 1.21 PFUs/ml, respectively. This lower reduction can be explained by the liquid nature of the virus preparations, which necessitated placing them horizontally on the surface for more indirect exposure, as well as the potential UV shielding caused by the viral suspension itself. To address these confounders, VSV was used as a virus sample which could be dried on a surface for an extended period and still retain its infectivity. This allowed the samples to be placed at 45 degrees for more direct exposure. Complete elimination of any detectable virus was noted in this case. It should be noted that viral contaminants would be in the dry form in most environmental situations in which PX-UV would be used.

Declaration

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The tests demonstrated that there was total elimination of the organisms at one and 15 minutes, respectively, for both the live Ebola virus and the *B. anthracis*. This indicates the potential of the PX-UV system to be deployed in an outbreak or biopreparedness manner.

The tests used here demonstrate the influence of laboratory methods over the reported effectiveness of technologies. For example, a large difference between wet and dry viral inocula was noted. A number of variables have been reported to influence the laboratory results in other research, including humidity, temperature, the smoothness of surfaces, protein loading, reflectivity, distance and other variables. While proof-of-concept data have been demonstrated in laboratory studies, readers are encouraged to seek data on the impact of a technology, such as PX-UV, in a real-world setting.[11,13–16]

Lastly, the organisms chosen for these tests represent organisms which were causing, or which would have the potential to cause, significant public health threats, including extended-spectrum beta lactamase-producing organisms, carbapenemase-producing organisms, resistant strains of A. baumannii and *S. aureus*, the Ebola virus and *B. anthracis*. The magnitude of the log reduction observed from the use of PX-UV against each of these organisms indicated that PX-UV disinfection played a role in preventing transmission of these organisms, and thereby in reducing the associated morbidity and mortality. Furthermore, by decreasing the probability of transmission of these organisms through a contaminated environment, PX-UV can contribute to efforts to address antimicrobial resistance by reducing the number of infections, and therefore extending the effectiveness of antibiotics by reducing demand for them.

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