Disinfection efficacy of multi-purpose disinfection solutions against ESKAPE pathogens and *Stenotrophomonas maltophilia*

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Introduction

The Centre for Disease Control (CDC) has identified the ESKAPE pathogens as an emerging threat to public health. These six strains of bacteria: *Enterococcus faecium*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Acinetobacter baumannii*, *Pseudomonas aeruginosa* and *Enterobacter* spp. are virulent and increasingly antibiotic resistant causing a high percentage of nosocomial infections in the United States. Additionally, *Stenotrophomonas maltophilia* has become an important ocular pathogen. The ISO standard for disinfection requires testing using non-resistant strains of *P. aeruginosa* and *S. aureus* only, resulting in a gap between disinfectant testing for regulatory claims and disinfectant testing against environmentally relevant resistant pathogens. This work presents suspension testing using non-standard organisms in three different multi-purpose disinfecting systems (MPDS), in an ISO accredited laboratory, to understand how MPDS perform against a selection of these pathogens.

Methodology

Bacterial isolates were cultured on relevant media at 37°C overnight. Inoculum suspensions were prepared at 1 x 10^8 $CFUmL^{-1}$ and diluted to a final concentration of 1 x 10^6 $CFUmL^{-1}$ in MPDS (Product A, Product B or Product C) or Phosphate Buffered Saline (PBS, Negative control). Following 3 or 6 hours treatment (50% and 100% of the relevant soak time) suspensions were neutralised with Dey-Engley broth for 5 minutes. Remaining viable organisms were enumerated by serial dilutions and spread plating. Tests were carried out in triplicate and the average and standard deviation calculated.

Results

At 0 hours, average negative control recoveries of 5.11 ± 0.05 $Log_{10}CFUmL^{-1}$, 5.32 ± 0.12 $Log_{10}CFUmL^{-1}$, 5.06 ± 0.06 $Log_{10}CFUmL^{-1}$, 5.39 ± 0.04 $Log_{10}CFUmL^{-1}$, 5.80 ± 0.09 $Log_{10}CFUmL^{-1}$ and 5.84 ± 0.09 $Log_{10}CFUmL^{-1}$ were observed for *S. aureus*, *A. baumannii*, *E. faecium*, *S. maltophilia, K. pneumoniae* and *P. aeruginosa*, respectively. Following 6 hours incubation with Product A, greater than 4 Log reductions were observed for *S. aureus, A. baumannii*, *S. maltophilia, K. pneumoniae* and *P. aeruginosa* (Table 1, Figure 1, Figure 2). Following 6 hours incubation with Product B, greater than 4 Log reductions were observed for all six species tested (Table 2 , Figure 1, Figure 2). Following 6 hours incubation with Product C, greater than 4 Log reductions were observed for all six species tested (Table 3 , Figure 1, Figure 2).

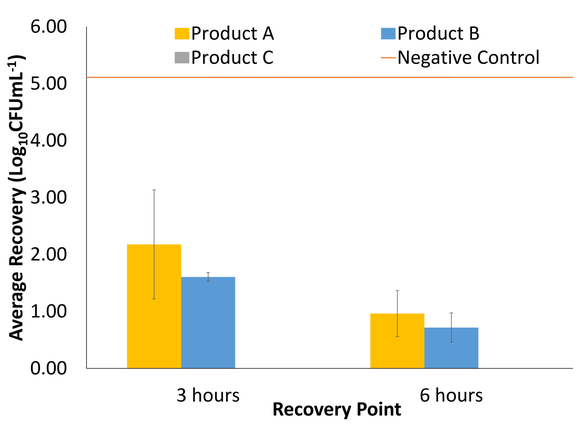

Figure 1. Average Log recoveries of *Staphylococcus aureus* compared to the negative control at 0 hours following 3 or 6 hours treatment with Product A, Product B or Product C.

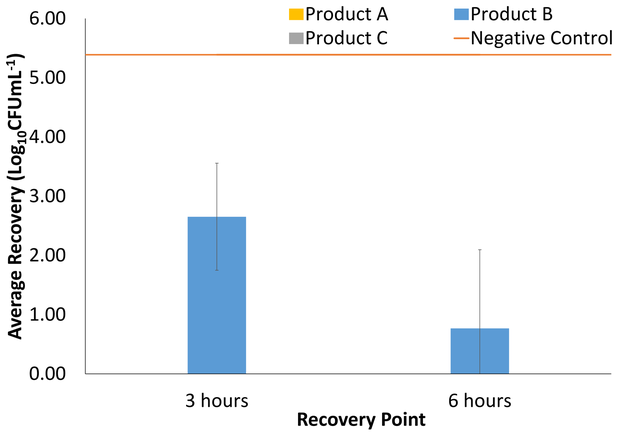

Figure 2. Average Log recoveries of *Stenotrophomonas maltophilia* compared to the negative control at 0 hours following 3 or 6 hours treatment with Product A, Product B or Product C.

Organism	Average Log Reduction ($Log_{10}CFUmL^{-1}$)	
	3 hours	6 hours
S. aureus	2.93	4.14
A. baumannii	5.32*	5.32*
E. faecium	2.64	3.65
S. maltophilia	5.39*	5.39*
K. pneumoniae	5.41	5.80*
P. aeruginosa	5.40	5.84*

Table 1. Average Log reductions of test organisms compared to the negative control at 0 hours following 3 or 6 hours treatment with Product A. * indicates no viable organisms recovered.

Organism	Average Log Reduction ($Log_{10}CFUmL^{-1}$)	
	3 hours	6 hours
S. aureus	3.50	4.39
A. baumannii	3.81	4.53
E. faecium	5.06*	5.06*
S. maltophilia	2.73	4.62
K. pneumoniae	5.80*	5.80*
P. aeruginosa	5.84*	5.84*

Table 2. Average Log reductions of test organisms compared to the negative control at 0 hours following 3 or 6 hours treatment with Product B. * indicates no viable organisms recovered.

Organism	Average Log Reduction ($Log_{10}CFUmL^{-1}$)	
	3 hours	6 hours
S. aureus	5.11*	5.11*
A. baumannii	4.85	5.32*
E. faecium	5.06*	5.06*
S. maltophilia	5.39*	5.39*
K. pneumoniae	5.80*	5.80*
P. aeruginosa	5.84*	5.84*

Table 3. Average Log reductions of test organisms compared to the negative control at 0 hours following 3 or 6 hours treatment with Product C. * indicates no viable organisms recovered.

Concluding comments and Discussion

Typically *Staphylococcus aureus* and *Pseudomonas aeruginosa* are the organisms of choice for standard test methods. Testing products against a range of Gram positive and Gram negative organisms may differentiate between products. Several ESKAPE species were chosen for this study to assess activity of MPDS against some of the most serious threats to human health. Treatment with Product C resulted in a greater than 4 Log reduction against all organisms tested, both at 6 hours incubation (recommended) and at the shorter 3 hours incubation (50% of recommeneded time). Treatment with Products A and B against two non-resistant organisms may have given a false reassurance which may then not be realised testing against environmental organisms, especially when dealing with resistance.

Testing against a larger range of bacterial species can differentiate between products. The species tested in this study have been chosen to represent some of the most serious pathogens and a species known to cause ocular infections. Testing a range of resistant organisms rather than the standard recommended organisms is more representative of the clinical scenario.

This project was carried out by Perfectus Biomed Group
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