Health Protection Scotland

NHS National Services Scotland

# Best methods for removal and destruction of pathogens

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## Contents

# Introduction

Over the past three years, Health Protection Scotland has carried out literature reviews on the following existing and emerging decontamination technologies of interest to the healthcare infection control community:

- Ultraviolet light (UV)
- Antimicrobial copper and silver solutions
- Chlorine dioxide
- High-intensity, narrow-spectrum (HINS) light
- Airborne hydrogen peroxide (HPV)
- Ozone
- Steam
- Wipes
- Microfibre
- Electrolysed water

The scope of this work included an assessment of effectiveness, as well as consideration of the practical and safety issues and costs related to their use in the healthcare environment. Recommendations on the use of these technologies in NHSScotland were made on the basis of the evidence identified; however they were not made with reference to specific pathogens.

This paper brings together the studies included in the existing and emerging decontamination technologies reviews, and presents a brief summary of the study results with respect to specific pathogens of relevance to the healthcare setting. This re-framing of the previously identified studies is intended to facilitate the development of recommendations for the best methods of pathogen removal and destruction by presenting the results in such a way as to allow the quality and quantity of studies and results for each technology to be easily determined at pathogen level.

# Methodology

All of the studies included in the ten existing and emerging decontamination technology reviews were assessed to determine whether they reported on the effectiveness of the technology in question with respect to specific pathogens. Studies that did not report on specific pathogens (e.g. studies that reported total viable count only) were excluded. Data extraction forms completed by the original reviewer were obtained for each included study. The reviewer produced a brief summary of the results of each study by referring to both the existing data extraction forms and the original study paper. The SIGN grading[1] and McDonald-Arduino evidence level[2], as determined by the original reviewer of each study was also collated. Graded recommendations were developed as appropriate.

# Results

A total of 74 studies from the ten existing and emerging technology reviews were relevant to this work. The results are presented in the table presented in **Appendix 1**. Columns are ordered with bacterial pathogens in alphabetical order first, followed by fungal then viral pathogens. Rows highlighted in grey are either inappropriate for terminal/supplementary cleaning e.g. detergent wipes, or are decontamination technologies that were not recommended for use in NHSScotland on the basis of the evidence identified in the existing and emerging technology reviews.

The majority of the included studies are graded as SIGN **level 3** evidence. Only two studies achieved a higher SIGN grading, both of which were graded as SIGN **level 2+** evidence.[3;4] There is greater variation in the grading of the studies using the McDonald-Arduino Evidentiary Hierarchy for Environmental Infection Control[2], with ten studies[5-14] (14%) achieving **level V** grading, which represents the strongest evidence for effectiveness: demonstrate reduced infections. However, the majority of studies[15-59] (45 studies: 60%) achieved a **level I** grading using the McDonald-Arduino hierarchy, which represents the weakest evidence for effectiveness: Laboratory demonstration of bioburden reduction efficacy. Seventeen studies[3;4;32;48;60-72] (23%) achieved a **level II** grading using the McDonald-Arduino hierarchy: demonstrate in-use bioburden reduction. One study[73] achieved a **level III** grading (demonstrate that in-use bioburden reduction may be clinically relevant), and another study[74] achieved a **level IV** grading (demonstrate reduced pathogen transmission via admission-discharge active surveillance testing or clinical incidence).

# Discussion

For the majority of pathogens listed in the table in **Appendix 1**, the quantity and/or quality of evidence on emerging and existing technologies is too low to make evidence-based recommendations.

However, there are a number of pathogens listed in the table (*Bacillus anthracis*; *Clostridium difficile*; *Escherichia coli*; *Enterococcus spp*., including VRE; *Pseudomonas aeruginosa*; *Staphylococcus aureus*, including MRSA; and Gram negative bacilli) with a greater quantity and/or quality of evidence on emerging and existing technologies.

## *Bacillus anthracis*

Seven studies assessed the efficacy of chlorine dioxide gas against *Bacillus anthracis* spores (or *Bacillus anthracis* spore surrogates: *Bacillus atrophaes* spores; *Bacillus subtillis* spores).[30;32;46;47;49;67;68] All seven studies were graded as SIGN **level 3** evidence, two of which were graded as **level II** [67;68] on the McDonald-Arduino hierarchy, and five were graded as **level I**[30;32;46;47;49]. This represents evidence at the lower end of the McDonald-Arduino evidentiary hierarchy, however *B.anthracis* is not a healthcare-associated pathogen, and as such it would be extremely unlikely that a study could be conducted that would achieve a higher score within that hierarchy.

The included studies all had different experimental protocols, including different pathogen inoculum concentrations, different environmental surfaces, different chlorine dioxide gas concentrations, different exposure times, and different ambient conditions (e.g. relative humidities). This makes it challenging to combine the results, and draw meaningful conclusions about the efficacy of chlorine dioxide gas against *Bacillus anthracis* spores. The majority of the studies report that >6 $\log_{10}$ reductions were achieved (on some surface materials); however there is variety across the studies in the concentrations and exposure times required for this level of reduction. In two studies, 6 $\log_{10}$ reductions were not achieved on specific surface materials at the concentrations and exposure times tested[30;46], and one study demonstrated that the dose required for complete inactivation was a function of material type.[47] In summary, the evidence indicates that chlorine dioxide gas can effectively inactivate *Bacillus anthracis* spores, but efficacy depends on the level of contamination, chlorine dioxide dose (concentration and exposure time) and surface material, and may also be influenced by ambient conditions.

Three studies assessed the efficacy of ozone against *Bacillus anthracis* spore surrogates (*Bacillus subtillis* spores; *Bacillus cereus* spores).[16;39;52;59] All four studies were graded as SIGN **level 3** evidence, and **level I** evidence on the McDonald-Arduino hierarchy. One study tested a combination of ozone and hydrogen peroxide, and it is not possible to determine the contribution of ozone alone to spore inactivation from this study.[59] Ozone concentrations tested ranged from 25pppm to 1500ppm, with exposure times of 20 minutes to 4 hours in the remaining three studies. There is no obvious dose-response relationship in terms of spore log reduction on the basis of these studies, with log reductions ranging from 1.27 to >3.1. In summary, there is a small volume of low

quality evidence on the efficacy of ozone against *Bacillus anthracis* spores. This evidence does not support the use of ozone for the inactivation of *Bacillus anthracis* spores.

### *Clostridium difficile*

A relatively large body of evidence was identified for the effectiveness of existing and emerging technologies against *C.difficile*.

Five studies assessed the effectiveness of airborne hydrogen peroxide decontamination systems against *C.difficile*.[9;13;28;60;62] All five studies were graded as SIGN **level 3** evidence. One study was graded as **level I**[28], two studies were graded as **level II**[60;62], and two studies achieved a **level V** grade[9;13] (representing the highest level of evidence) on the McDonald-Arduino hierarchy. Both studies graded as level V evidence demonstrated reduced risk of *C.difficile* infection in patients admitted to rooms decontaminated with HPV compared to patients admitted to rooms decontaminated using standard terminal cleaning methods; however the reduction was only significant in one study.[9] Both of the studies that achieved a level II grade compared HPV to hypochlorite solution (5,000ppm; 1,000ppm), and found HPV to be more effective against *C.difficile* spores.[60;62] The study that was graded as level I evidence demonstrated >6 $\log_{10}$ reduction in *C.difficile* spores after 20 minutes exposure to 100,000ppm HPV. This evidence suggests that HPV may be effective against C*.difficile* spores, and could be a useful adjunct to standard terminal cleaning methods.

Eight studies assessed the efficacy of wipes against *C.difficile*.[5;45;48;53;62;65;66;74] All eight studies were graded as SIGN **level 3** evidence. Three studies were graded as **level I**[45;48;53], three studies were graded as **level II**[62;65;66], one study was graded as **level IV**[74], and one study was graded as **level V**[5] on the McDonald-Arduino hierarchy. The studies tested a range of different wipe formulations using different study protocols, making it challenging to draw meaningful conclusions. Two of the Level I studies demonstrated the limited availability of the detergent wipes and disinfectant wipes tested (17 different wipes in total: 10 disinfectant and 7 detergent wipes) to physically remove spores. Both of these studies demonstrated that wipes (16/17) failed to retain spores, transferring them between surfaces.[45;53] The other level I study demonstrated that disinfectant wipes (hydrogen peroxide + peracetic acid; sodium hypochlorite) were effective in removing and inactivating spores.[48] One level II study found that none of the four different disinfectant wipes tested were able to eliminate *C.difficile* contamination on a tablet computer, and none demonstrated residual effect.[66] Another level II study demonstrated that peracetic acid wipes were less effective against *C.difficile* spores than 1,000ppm hypochlorite solution.[62] The remaining level II study only one of the five disinfectant wipes performed better than gauze and water at removing spores from the surface of an anaesthetic machine (0.55% sodium hypochlorite wipe), and none of the wipes performed better than gauze and water at removing spores from flat and ridged caps (intended to simulate machine knobs).[65] Both the level IV and the level V study demonstrated reduced *C.difficile* infection rates in patients when a wipe based cleaning regimen (0.55% active chlorine wipes, and peracetic acid wipes respectively) was in place, relative to when a quaternary ammonium compound/chlorine based cleaning regimen respectively was in place.[5;74] However, in one study, other interventions to reduce *C.difficile* infection rates were introduced simultaneously with the wipe based cleaning regimen, so it is not possible to attribute the reduction in infection rates to the use of wipes.[5] Overall, the evidence on the efficacy of wipes against *C.difficile* is mixed and inconclusive, in part due to the variety of different wipe formulations available. There is some evidence that detergent and disinfectant wipes can transfer spores between surfaces, emphasising the importance of using one wipe for one surface.

Five studies assessed the efficacy of UV light decontamination systems against *C.difficile*.[3;8;10;12;14] Four of these studies were graded as SIGN **level 3** evidence[8;10;12;14], and one was graded as SIGN **level 2+** evidence.[3] One study was graded as **level II** evidence[3] on the McDonald-Arduino hierarchy, with the remaining four studies achieving a **level V** grading, representing the highest level of evidence.[8;10;12;14] Four of the studies tested pulsed xenon UV light

systems[3;8;10;14], while one tested a UV-C system.[12] Three of the studies that assessed pulsed xenon UV systems demonstrated reductions in *C.difficile* infection rates when UV disinfection was used as an adjunct to standard and/or terminal cleaning. However, in two of these studies other interventions to reduce *C.difficile* infection rates were introduced at the same time, so it is not possible to attribute the reduction in infection rates to the use of pulsed xenon UV.[8;10] The other study that assessed a pulsed xenon UV disinfection system found that decontamination of hospital rooms with hydrogen peroxide disinfectant followed by UV was equivalent to decontamination with hydrogen peroxide solution followed by 10% sodium hypochlorite solution in terms of reducing environmental *C.difficile* contamination.[3] The study that assessed a UV-C disinfection system demonstrated a significant reduction in *C.difficile* infection rates when UV-C was added to the terminal cleaning regimen.[12] In summary, there is some evidence to suggest that the use of UV disinfection systems as an adjunct to terminal cleaning may reduce environmental contamination with *C.difficile*, and thus reduce *C.difficile* infection rates.

Three studies assessed the efficacy of chlorine dioxide solutions against *C.difficile*.[7;43;55] All three studies were graded as SIGN **level 3** evidence. Two of the studies were graded as **level I** evidence[43;55], and one achieved a **level V** grade on the McDonald-Arduino hierarchy.[7] One of the level I studies demonstrated that long contact times (10-30 minutes) were required for >6 $\log_{10}$ reduction in *C.difficile* spores.[43] The other level I study demonstrated that only 8/16 different chlorine dioxide products tested achieved >3 $\log_{10}$ reduction in *C.difficile* spores in suspension after 1 minute contact time.[55] The level V study fund no impact on either *C.difficile* infection rates or on environmental contamination with *C.difficile* after a hospital-wide switch from routine cleaning with microfibre and water and enhanced cleaning with a 1,000ppm chlorine releasing agent to cleaning with a commercially available chlorine dioxide product for all routine and terminal cleaning.[7] Overall, the identified evidence for the efficacy of chlorine dioxide solutions against *C.difficile* is limited, and does not currently support its use.

### *Escherichia coli*

Six studies assessed the efficacy of high intensity narrow spectrum (HINS) light against *E.coli*, all of which were graded as SIGN **level 3** evidence and as **level I** evidence on the McDonald-Arduino hierarchy, representing the lowest level of evidence.[34;35;37;38;40;41] The studies tested different doses (J/cm$^2$), irradiance levels (W/cm$^2$), and exposure times, and demonstrated different log reductions, making it challenging to aggregate the results and draw meaningful conclusions. The results of the studies indicate that HINS light can inactivate *E.coli*, but the optimum combination(s) of dose, irradiance level and exposure time for complete inactivation cannot be determined on the basis of the evidence identified.

### *Enterococcus spp.*, including VRE

Four studies assessed the efficacy of UV light decontamination systems against Vancomycin resistant *Enterococci* (VRE).[8;12;14;72] All three studies were graded as SIGN **level 3** evidence. One study was graded as **level II** evidence[72], while the other three studies achieved a **level V** grade on the McDonald-Arduino hierarchy, representing the highest level of evidence.[8;12;14] Three studies tested pulsed xenon UV decontamination systems[8;14;72], while one study tested a UV-C decontamination system.[12] The two level V studies that tested pulsed xenon UV systems demonstrated a reduction in VRE infection rates when UV disinfection was used as an adjunct to terminal cleaning.[8;14] However, one study demonstrated a significant reduction in infections in ICU only (not at facility-wide level)[14], and in the other study additional interventions to reduce multidrug resistant organism infection rates were introduced at the same time, so it is not possible to attribute the reduction in infection rates to the use of pulsed xenon UV.[8] The other study that tested a pulsed xenon UV system demonstrated that use of the system as an adjunct to terminal cleaning of rooms that had been occupied by VRE patients eliminated residual environmental contamination with VRE following terminal cleaning.[72] The study that assessed a UV-C decontamination system, demonstrated a non-significant reduction in VRE infection rates when the system was

used as an adjunct to terminal cleaning.[12] Overall, the evidence is limited, but suggests that the use of UV disinfection systems as an adjunct to terminal cleaning may reduce environmental VRE contamination, and thus reduce VRE infection rates.

Three studies assessed the efficacy of disinfectant wipes against VRE.[19;66;75] All three studies were graded as SIGN **level 3** evidence. Two studies were graded as **level II** evidence[66;75], and one was graded as **level I** evidence on the McDonald-Arduino hierarchy.[19] The studies tested a range of different wipe formulations (with some overlap between studies) using different study protocols, making it challenging to draw meaningful conclusions. Both of the level II studies tested a range of wipes against artificially contaminated computer hardware (tablet computer and keyboard).[66;75] One study found that all four of the wipes tested were significantly better at removing VRE contamination from a tablet computer than the manufacturer recommended lint free cloth. The study demonstrated that three of the wipes tested (Chlorox wipe; Sani-Cloth CHG 2%; Tristel Sporicidal) were the most effective, removing most of the VRE from the tablet surface, and one wipe (Sani-Cloth) demonstrated a residual effect (up to 12 hours).[66] The other level II study demonstrated an average removal of between 98.71 and 100% CFU from a computer keyboard after a 5-second wipe for the 5 different wipes tested (a sterile water wipe demonstrated average removal of 99.61% CFU, indicating that physical removal of spores without disinfection is important). The three quaternary ammonium compound-based wipes (Sani-Cloth Plus; Chlorox; CaviWipes) demonstrated 100% residual efficacy (48 hours) against VRE after a 10 minute contact time.[75] The level I study found no difference in efficacy against VRE between the 5 different wipes tested when wipes were swiped across a contaminated plastic surface 3 or 5 times.[19] In summary, there is a small volume of evidence suggesting that disinfectant wipes may be appropriate for decontamination of VRE contaminated surfaces that cannot be decontaminated with hypochlorite solution (e.g. tablet computers), however the evidence identified is insufficient to identify the most effective wipe formulation(s) or wiping protocol.

### *Pseudomonas aeruginosa*

Three studies assessed the efficacy of high intensity narrow spectrum (HINS) light against *P.aeruginosa,* all of which were graded as SIGN **level 3** evidence and as **level I** evidence on the McDonald-Arduino hierarchy, representing the lowest level of evidence.[35;37;38] The studies tested different doses ($J/cm^2$), irradiance levels ($W/cm^2$), and exposure times, and demonstrated different log reductions, making it challenging to aggregate the results and draw meaningful conclusions. The results of the studies suggest that HINS light may be able to inactivate *P.aeruginosa,* but the optimum combination(s) of dose, irradiance level and exposure time for complete inactivation cannot be determined on the basis of the evidence identified.

### *Staphylococcus aureus*, including MRSA

The largest body of evidence was identified for the effectiveness of existing and emerging technologies against *Staphylococcus aureus,* including MRSA.

Six studies assessed the efficacy of high intensity narrow spectrum (HINS) light against *Staphylococcus aureus*, all of which were graded as SIGN **level 3** evidence. One study was graded as **level II** evidence[69], and the remaining five were graded as **level I** evidence on the McDonald-Arduino hierarchy, representing the lowest level of evidence.[34-38] The studies tested different doses ($J/cm^2$), irradiance levels ($W/cm^2$), and exposure times, and demonstrated different log reductions, making it challenging to aggregate the results and draw meaningful conclusions. The results of two level I studies suggest that higher doses may be required for inactivation of MRSA than for inactivation of MSSA.[34;35] Overall, the results indicate that HINS light can inactivate *Staphylococcus aureus,* but the optimum combination(s) of dose, irradiance level and exposure time for complete inactivation cannot be determined on the basis of the evidence identified.

Four studies assessed the efficacy of airborne hydrogen peroxide decontamination systems against *Staphylococcus aureus*.[11;13;63;73] All four studies were graded as SIGN **level 3** evidence. One study was graded as **level II** evidence[63], one study was graded as **level III** evidence[73], and two studies achieved a **level V** grading on the McDonald-Arduino hierarchy, representing the highest level of evidence.[11;13] Both studies graded as level V evidence demonstrated reduced risk of MRSA acquisition in patients admitted to rooms where HPV decontamination was used as an adjunct to terminal cleaning. Only one of these studies demonstrated a significant reduction, however this study has major limitations: the comparison was with detergent-only terminal cleaning and other interventions to reduce MRSA acquisition rates were introduced at the same time, meaning that it is not possible to attribute the reduction to the use of HPV.[11] The level III study demonstrated that HPV decontamination of a hospital room vacated by a patient with history of MRSA infection and colonisation following terminal cleaning further reduced, but did not eliminate, MRSA environmental contamination.[73] The level II study compared manual terminal cleaning to HPV decontamination of rooms vacated by MRSA positive patients, and found that MRSA contamination was lower in the rooms subject to HPV decontamination. However, statistical analysis was not reported, and detergent-only was used for terminal cleaning, which is not a standard terminal cleaning regimen within NHSScotland.[63] In summary, the evidence on the effectiveness of HPV against *S.aureus* is subject to methodological limitations, and is insufficient to recommended HPV as an adjunct to terminal cleaning in the context of *S.aureus* contamination.

Four studies assessed the efficacy of UV decontamination systems against *Staphylococcus aureus*.[4;8;12;14] Three studies were graded as SIGN **level 3** evidence[8;12;14], and one was graded as SIGN **level 2+** evidence.[4] One study was graded as **level II** evidence[4], and three studies were graded as **level V** on the McDonald-Arduino hierarchy, representing the highest level of evidence.[8;12;14] Three studies tested pulsed xenon UV decontamination systems[4;8;14], while one study tested a UV-C decontamination system.[12] The two level V studies that tested pulsed xenon UV systems demonstrated a reduction in MRSA infection rates when UV disinfection was used as an adjunct to terminal cleaning.[8;14] However, in one of these studies the reduction at the facility-wide and ICU-level was not significant, and there was actually a (non-significant) increase in rates in non-ICU areas[14], and in the other study additional interventions to reduce multidrug resistant organism infection rates were introduced at the same time, so it is not possible to attribute the reduction in infection rates to the use of pulsed xenon UV.[8] The other study that tested the efficacy of a pulsed xenon UV disinfection system against MRSA demonstrated that it was significantly better than manual cleaning of high touch surfaces with hypochlorite solution.[4] The study that assessed a UV-C decontamination system, demonstrated a non-significant reduction in MRSA infection rates when the system was used as an adjunct to terminal cleaning.[12] Overall, the evidence is limited, but suggests that the use of UV disinfection systems as an adjunct to terminal cleaning may reduce environmental MRSA contamination, which may have an impact on MRSA infection/colonisation rates.

Seven studies assessed the effectiveness of wipes (detergent and/or disinfectant) against *S.aureus*.[19;45;51;57;65;66;75] All seven studies were graded as SIGN **level 3** evidence. Four of these studies were graded as **level I** evidence[19;45;51;57], and three were graded as **level II** evidence on the McDonald-Arduino hierarchy.[65;66;75] The studies tested a range of different wipe formulations (with some overlap between studies) using different study protocols, making it challenging to draw meaningful conclusions. Two of the level II studies tested a range of wipes on artificially contaminated computer hardware (tablet computer and keyboard)[66;75], and one tested a range of wipes on an artificially contaminated anaesthesia machine and flat and ridged caps (to simulate machine knobs).[65] One level II study found that all four of the wipes tested were significantly better at removing MRSA contamination from a tablet computer than the manufacturer recommended lint free cloth. The study demonstrated that three of the wipes tested (Chlorox wipe; Sani-Cloth CHG 2%; Tristel Sporicidal) were the most effective, removing most of the MRSA from the tablet surface, and one wipe (Sani-Cloth) demonstrated a residual effect (up to 12 hours).[66] Another level II study demonstrated an average removal of between 95.29 and 100%

CFU from a computer keyboard after a 5-second wipe for the 5 different wipes tested (a sterile water wipe demonstrated average removal of 98.58% CFU, indicating that physical removal of spores without disinfection is important).[75] The remaining level II study found that although all five disinfectant wipes tested removed *S.aureus* from the surface of the anaesthesia machine, their performance was not better than gauze with water. A 0.5% hydrogen peroxide wipe was most effective at removing *S.aureus* from flat and ridged caps, and one wipe was outperformed by gauze and water in the context of flat and ridged caps.[65] One of the level I studies found no difference in efficacy against MRSA between the 5 different disinfectant wipes tested when wipes were swiped across a contaminated plastic surface 3 or 5 times.[19] Another level I study demonstrated the limited availability of the detergent wipes (7 different wipes) to physically remove *S.aureus*. [45] Another level I study tested both detergent and disinfectant wipes (two different wipes), and found that they demonstrated similar performance in terms of removing the pathogen from clean and dirty surfaces. The study found that detergent wipes demonstrated bactericidal effect, while detergent wipes did not.[57] The final level I study found that three of the five most effective disinfectant wipes tested resulted in ≥ 7 $\log_{10}$ reduction (two accelerated $H_2O_2$ wipes; 1 sodium hypochlorite wipe with 1000ppm available chlorine), and the least effective resulted in >4 $\log_{10}$ reduction.[51] Three of the level I studies found that some or all of the wipes tested (11/14 overall) transferred *S.aureus* to a subsequent surface or surfaces.[45;51;57] Overall, the evidence on the efficacy of wipes against *S.aureus* is mixed and inconclusive, in part due to the variety of different wipe formulations available. There is a small volume of evidence to suggest that disinfectant wipes may be appropriate for decontamination of *S.aureus* contaminated surfaces that cannot be decontaminated with hypochlorite solution (e.g. tablet computers), however the evidence identified is insufficient to identify the most effective wipe formulation(s) or wiping protocol. There is some evidence that detergent and disinfectant wipes can transfer spores between surfaces, emphasising the importance of using one wipe for one surface.

### Gram negative bacilli

Four studies assessed the effectiveness of airborne hydrogen peroxide decontamination systems against Gram-negative rods.[13;61;71;73] All four studies were graded as SIGN **level 3** evidence. Two studies were graded as **level II** evidence[61;71], one was graded as **level III** evidence[71], and one was graded as **level V** evidence on the McDonald-Arduino hierarchy.[13] Both of the level II studies and the level III study compared the number of environmental samples that tested positive for Gram negative rods after terminal/deep cleaning, then after subsequent HPV decontamination in hospital wards.[61;71;73] The study that compared HPV decontamination with deep cleaning (2,000ppm sodium hypochlorite, and 70% alcohol wipes for equipment) on the presence of gram negative rod environmental contamination in an ICU demonstrated that the proportion of positive samples reduced from 47.6% after deep cleaning to zero following HPV decontamination.[71] The other level II study demonstrated a significant reduction in the number of samples that tested positive after HPV decontamination compared to after standard terminal cleaning alone.[61] The level III study demonstrated that HPV decontamination of a hospital room vacated by a patient with history of gentamycin resistant Gram negative rod infection and colonisation reduced the number of positive samples following terminal cleaning from 10% to zero.[73] The level V study demonstrated a reduced risk of acquisition of Gram negative rods (individually) in patients admitted to rooms decontaminated with HPV, compared to rooms decontaminated using standard methods, however this was not significant.[13] In summary, the evidence on the effectiveness of HPV against Gram-negative rods is limited, but suggests that it may be effective. However, the evidence is currently insufficient to recommended HPV as an adjunct to terminal cleaning in the context of Gram negative rod contamination.

## Conclusion

Despite the volume of evidence identified overall, few recommendations can be drawn for the best methods for pathogen removal. This is in part due to the low volume of evidence for specific technologies. In addition, where a body of evidence was identified on a specific technology for a particular pathogen, studies used different protocols, including e.g. different product concentrations, formulations and contact times, making it challenging to make comparisons and draw meaningful conclusions. Practical and cost considerations associated with use of the technologies included in this review in healthcare settings must be considered.

## Recommendations

This review makes the following recommendations:

- HPV or UV disinfection systems may be effective against C*.difficile* spores, and could be a useful adjunct to standard terminal cleaning methods. **(Grade D Recommendation)**
- The use of UV disinfection systems as an adjunct to terminal cleaning may reduce environmental VRE contamination, and thus reduce VRE infection rates. **(Grade D Recommendation)**
- The use of UV disinfection systems as an adjunct to terminal cleaning may reduce environmental MRSA contamination, which may have an impact on MRSA infection/colonisation rates. **(Grade D Recommendation)**

# Appendix 1: Table presenting a summary of existing and emerging technologies studies by pathogen

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
|---|---|---|---|---|---|
| ***Acinetobacter spp.*** | *Acinetobacter calcoaceticus Acinetobacter baumanii* | AM copper (3 different formulas) | 2-3 $log_{10}$ reduction at 1ppm<br>>6 $log_{10}$ reduction in 60 mins at 150ppm<br>All three formulas (on an ultra microfibre cloth) at a concentration of 150ppm removed an initial inoculum of 2 x $10^6$ CFU. No CFU were recovered from the cloth after 16 hours. Ultra microfibre cloth and water also removed the inoculum, however CFU were recovered from the cloth after 16 hours. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Gant et al 2007[24] |
| | *Acinetobacter baumanii* | Chlorine dioxide gas | Complete inactivation in ¾ decontamination trials:<br>• 362ppm, 850ppm-hours, 55% RH;<br>• 406ppm, 763ppm-hour, 65% RH;<br>• 695ppm, 756ppm hours, 55% RH. | SIGN **level 3** evidence<br>McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Lowe et al 2013b[67] |
| | *Acinetobacter baumanii* | Chlorine dioxide gas | Complete inactivation at:<br>• 351ppm, 667ppm-hours, 50% RH;<br>• 377ppm, 890ppm-hours, 65% RH;<br>• 379ppm, 767ppm-hour, 65% RH;<br>• 385ppm, 770ppm-hour, 65% RH. | SIGN **level 3** evidence<br>McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Lowe et al 2013c[76] |
| | *Acinetobacter baumanii* | Chlorine dioxide liquid | >6 $log_{10}$ reduction after 5 minutes contact time at 22 ± 2°C for both 500ppm and 1000ppm. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Sabbah et al 2010[50] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
|---|---|---|---|---|
| *Acinetobacter baumanii* | HINS | 180 minute exposure for 4.2 $log_{10}$ reduction | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | MacLean et al 2009[35] |
| Imipenem resistant *Acinetobacter baumanii* | HPV | All *A.baumanii* contamination in all rooms tested removed by terminal cleaning alone, so HPV not necessary for removal of this organism. [Only 0.41% of samples contaminated with *A.baumanii* after patient discharge, so there was a low level of initial contamination.] | SIGN **level 3** evidence McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Blazejewski et al 2015[61] |
| *Acinetobacter baumanii* | Ozone | 25ppm ozone for 20 minutes followed by a short burst of humidity >90% resulted in ≥4 $log_{10}$ reduction | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Sharma and Hudson 2008[52] |
| *Acinetobacter baumanii* | Steam – dry steam | 4.9 $log_{10}$ reduction after 5 seconds | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Song et al 2012[54] |
| XDR-*Acinetobacter baumanii* | Steam – dry steam | Kill-time 5 minutes for inoculum with or without bovine serum albumen (initial inoculum $10^9$ CFU/mL). 5 minute kill time for 2.8% sodium hypochlorite with same inoculum concentration in the absence of BSA, but longer kill time required in presence of BSA (i.e. organic soil). | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Bagatinni et al 2015[17] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
|---|---|---|---|---|
| *Acinetobacter baumanii* | UV-C | Reduction in *A.baumanii* infection incidence rates from 0.39 in the baseline period (terminal cleaning only) to 0.11 in the intervention period (terminal cleaning + UV-C disinfection) (incidence rate change -71.8%, p=0.005). | SIGN **level 3** evidence McDonald-Arduino **Level V** – Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | Napolitano et al 2015[12] |
| *Acinetobacter baumanii* | Detergent wipes (7 different wipes) | Average removal of *A.baumanii* CFU from a stainless steel surface after a 10-second wipe was 3.51$\log_{10}$ (range 3.01-3.81). There was no statistically significant difference between the 7 wipes tested.<br>Wipe C (Sani Cloth Detergent Wipe) performed worst at preventing subsequent transfer to a stainless steel surface, with 8.05% transfer. All other wipes transferred less than 0.1%.<br>[Wipes performed better for removal and prevention of transfer for *A.baumanii* than for the other organisms tested (i.e. *S.aureus* and *C.difficile* spores).] | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Ramm et al 2015[45] |
| *Acinetobacter baumanii* | Disinfectant Wipes (5 different wipes) | The three most effective wipes (2 0.5% accelerated H2O wipes; 1 sodium hypochlorite wipe with 1000ppm available chlorine) resulted in at least 7 $\log_{10}$ CFU reduction, and the least effective wipe resulted in >5 $\log_{10}$ reduction after a 10-second wipe. Only one wipe resulted in subsequent transfer to another stainless steel surface. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Sattar et al 2015[51] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| ***Bacillus anthracis*** | *Bacillus anthracis* spores and vegetative cells | Chlorine dioxide gas | Complete activation of spores and vegetative cells in 3/6 decontamination trials:<br>• 377ppm, 890ppm-hours, 65% RH;<br>• 379ppm, 767ppm-hours, 65% RH;<br>• 385ppm, 770ppm-hours, 65% RH.<br>Higher % of inactivation for vegetative cells (99%) than for spores (93-98%) in the other 3 trials.<br>[Initial inoculum $10^{10}$ CFU] | SIGN **level 3** evidence<br>McDonald-Arduino **Level II –** Demonstration of in-use bioburden reduction effectiveness | Lowe et al 2013a[68] |
| | *Bacillus anthracis* spores | Chlorine dioxide gas | Complete inactivation not achieved in any of the 4 decontamination trials (maximum exposure of 850ppm-hours). Inactivation ranged from 88-96%.<br>[Initial inoculum $10^{10}$ CFU] | SIGN **level 3** evidence<br>McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Lowe et al 2013b[67] |
| | *Bacillus anthracis* spores | Chlorine dioxide liquid | After 3 minutes exposure:<br>• 100% kill (8 $\log_{10}$ reduction) at 10.0mg/mL<br>• 99.99% kill (4.34$\log_{10}$ reduction) at 5.0mg/mL<br>• 97.3% kill (1.57 $\log_{10}$ reduction) at 2.5mg/mL | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Chatuev and Peterson 2010[21] |
| | *Bacillus anthracis* spores | Chlorine dioxide gas (1 wet system; 1 dry system) | There was a general trend in reduction in the number of viable spores with increased exposure time at each of the four chlorine dioxide concentrations tested (500; 1,000; 1,500; 3,000ppmv).<br>Kill-time was a function of dose (concentration multiplied by exposure time: ppmv-h).<br>Time required for 6 $\log_{10}$ reduction a function of material type: carpet, cinder block and ceiling tiles required 3,000 to 6,000ppmv-h; | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Rastogi et al 2010[47] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | | steel and wallboard required 6,000 to 9,000ppmv-h; and pinewood required >9,000ppmv-h. Cinder block was the only material where kill time was a function of concentration.<br>No difference in efficacy between the two methods (wet system and dry system). | | |
| | *Bacillus anthracis* spores | Chlorine dioxide gas | For a spore inoculum of 1 x $10^6$ exposed to chlorine dioxide gas at 3,600ppmv for 3 hours (10,800ppmv-h), log reduction values ranged between 2.5 for wood and 6.6 for ceiling tile.<br>No significant drop in log reduction at 1 x $10^7$ or 1 x $10^8$ inoculum concentration. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Rastogi et al 2009[46] |
| ***Bacillus atrophaeus*** **(Surrogate for *Bacillus anthracis* spores)** | *Bacillus atrophaeus* spores | Chlorine dioxide gas | Complete kill (at least 6 $\log_{10}$ reduction) for a dose of 720ppm-h (estimated 2-hr exposure time).<br>[A number of practical/logistical limitations are outlined in the paper.] | SIGN **level 3** evidence<br>**Level I** – Laboratory demonstration of bioburden reduction efficacy | Lowe et al 2012[32] |
| | *Bacillus atrophaeus* spores | Chlorine dioxide gas | Complete activation of spores in 4/6 decontamination trials:<br>• 351ppm, 677ppm-hours, 50% RH;<br>• 377ppm, 890ppm-hours, 65% RH;<br>• 379ppm, 767ppm-hours, 65% RH;<br>• 385ppm, 770ppm-hours, 65% RH.<br>[Initial inoculum $10^{10}$ CFU] | SIGN **level 3** evidence<br>McDonald-Arduino **Level II –** Demonstration of in-use bioburden reduction effectiveness | Lowe et al 2013a[68] |
| | *Bacillus atrophaeus* spores | Chlorine dioxide gas | Complete inactivation not achieved any of the 4 decontamination trials (maximum exposure 850ppm-hours). Spores remained viable in at least one of the nine test sites after each of the 4 decontamination trials.<br>[initial inoculum $10_{10}$ CFU] | SIGN **level 3** evidence<br>McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Lowe et al 2013b[67] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | *Bacillus atrophaeus* spores | Disinfectant wipes (5 different wipes) | Only 2/5 wipes performed better than gauze and water at removing spores from the smooth, flat surface of an anaesthetic machine: 0.55% sodium hypochlorite wipe; 0.5% hydrogen peroxide wipe.<br>None of the 5 wipes performed better than gauze and water at removing spores from flat and ridged caps, and one wipe performed significantly worse than gauze and water: 0.28%/17.2% Diisobutylphenoxyethoxyethyl dimethyl benzyl ammonium chloride wipe.<br>Authors concluded that wetness of wipes may play a role in effectiveness.<br>[*Bacillus atrophaeus* spores were more difficult to clean than *S.aureus*.] | SIGN **level 3** evidence<br>McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Gonzalez et al 2015[65] |
| ***Bacillus subtilis* spores (Surrogate for *Bacillus anthracis* spores)** | *Bacillus subtilis* spores | Chlorine dioxide gas | >6 $\log_{10}$ reduction on all materials (galvanised steel, carpet, wood, painted wallboard paper) after 6 hours exposure at 750ppmv (75% RH, 24°C). No detectable CFUs on any material after the maximum exposure time of 12 hours.<br>Actual time to achieve 6 $\log_{10}$ reduction was a function of material type and (depending on material) inoculation method (aerosol or liquid). Galvanised steel was the easiest to decontaminate, and wood was the most difficult. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Ryan et al 2014[49] |
| | *Bacillus subtilis* var. *niger* spores | Chlorine dioxide gas | Log reduction values ranged from 1.80 (cotton cloth) to 6.64 (glass) for the 6 materials tested (stainless steel, painted steel, polyvinyl chloride, polyurethane, glass, and cotton cloth) with an exposure dose of 0.080% chlorine dioxide gas for 3 hours. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Li et al 2012[30] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | Where materials were humidified prior to decontamination (i.e. exposed to 70-75% RH for 1 hour), log reduction ranged from 2.69 (cotton cloth) to 5.88 (glass).<br>There was a statistically significant difference between the sporicidal efficacy of chlorine dioxide between porous and nonporous materials. | | |
| *Bacillus subtilis* spores | Chlorine dioxide liquid | Initial inoculum of 100µL (~$10^8$ CFU/mL) exposed to stabilized chlorine dioxide at a concentration of 187µg/mL was completely inactivated after 10 min contact time. At 47µg/mL, inactivation was incomplete after 60 minutes contact time, but was complete after 24 hours. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Friedline et al 2015[23] |
| *Bacillus subtilis spores* | Chlorine dioxide liquid | Chlorine dioxide at a concentration of 630 ± 60 mg/L free chlorine achieved >6 $\log_{10}$ reduction (complete inactivation) of spores in Columbia broth in ~10 minutes, however for spores in brain heart infusion broth, ~30 min contact time was required for the same level of activity. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Perez et al 2005[43] |
| *Bacillus subtilis* spores | HINS | With exposure to HINS light (405nm) with an irradiance of 40mW/$cm^2$, there was ~0.6 $\log_{10}$ CFU/mL reduction with an exposure to a dose of 0.58kJ/$cm^2$, and a ~1.7 log10 CFU/mL reduction with an exposure of 1.15kJ/$cm^2$. Both reductions were significantly better than the control (no HINS).<br>No significant difference in inactivation rates between *Bacillus subtilis, Bacillus cereus and Bacillus megaterium.* | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | McLean et al 2013b[36] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | *Bacillus subtilis* spores | Ozone + hydrogen peroxide | In a test chamber, exposure to 80ppm ozone + 1% hydrogen peroxide or 80ppm ozone + 3% hydrogen peroxide at 80% RH resulted in a 7.23 $log_{10}$ reduction after 90 minutes exposure.<br>In a 113$m^3$ test room, exposure to 80ppm ozone + 1% hydrogen peroxide at 80% RH resulted in a 6.37 $log_{10}$ reduction after 90 minutes exposure. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Zoutman et al 2011[59] |
| | *Bacillus subtilis* spores | Ozone | Ozone at 3 mg/L (1500 ppm) produced ~3-log reduction within 4 hr at 90% RH and 22 °C on glass surfaces. The inactivation curves consisted of a short lag phase followed by an exponential decrease in the number of surviving spores.<br>No additional benefit was observed in terms of increased inactivation rate at higher ozone concentrations.<br>Higher humidity levels and pre-hydration of the spores increased the rate of inactivation using ozone gas. The type of surface on which the spores were had an impact on the rate of inactivation. Inactivation rates on glass, a vinyl floor tile, and office paper were nearly the same, whereas cut pile carpet and hardwood flooring surfaces resulted in much lower inactivation rates. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Aydogan and Gurol 2006[16] |
| ***Bacillus cereus* (Surrogate for *Bacillus anthracis*)** | *Bacillus cereus* vegetative cells | AM silver (5 different disinfectants, including 3 different silver nanoparticle solutions) | Agar diffusion tests were used to test activity of 3 different silver nanoparticle solutions: silver nanoparticle solution (NP; 6 mg/ml), concentrated silver nanoparticle solution (CNP; 60 mg/ml), and silver nanoparticle solution containing additional sodium chloride (NPNaCl) and two additional disinfectants | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Araujo et al 2012[15] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | dodecyltrimethylammonium bromide (Dotab), silver sulfadiazine (SAg). For *B. cereus*, there was no difference between the NP and CNP treatments, whereas the inhibition caused by NPNaCl was lower. There was no significant difference ($P > 0.05$) in bacterial inhibition by Dotab and SAg for *B. cereus.* A smaller inhibition was observed for Dotab and SAg treatments than for the others (NP, CNP, and NPNaCl).<br>Solutions (Dotab (0.0312 M), sodium carbonate (40 mg/L), and CNP (60 mg/L)) were also tested for their ability to remove or kill *B. cereus* cells adhered to stainless steel surfaces. CNP demonstrated the best performance ($p<0.05$). | | |
| *Bacillus cereus* spores and vegetative cells | HINS | Vegetative cells: There was an initial plateau with no significant inactivation, but after exposure to a dose of 48 J/cm$^2$, there was significant inactivation with near complete inactivation (4 $\log_{10}$ CFU/mL reduction) achieved after exposure to 108 J/cm$^2$. *B.cereus* more resistant to inactivation than *C.difficile*, with more than double the dose required for a similar log reduction.<br>Spores: With exposure to HINS light (405nm) with an irradiance of 40mW/cm$^2$, there was ~0.7 $\log_{10}$ CFU/mL reduction with an exposure to a dose of 0.58kJ/cm$^2$ (this was not significant), and a ~1.8 log10 CFU/mL reduction with an exposure of 1.15kJ/cm$^2$ (this was significantly better than the control).<br>Significantly greater doses were required for | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | MacLean et al 2013b[36] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | | inactivation of spores than vegetative cells. No significant difference in inactivation rates between *Bacillus subtilis, Bacillus cereus and Bacillus megaterium.* | | |
| | *Bacillus cereus* spores | Ozone | Inoculum: 20µL, 1 x $10^5$ CFU/ml. Exposure to 25ppm ozone for 30 min, with a 10 minute quench to remove residual ozone in a laboratory setting resulted in a 1.27 $log_{10}$ reduction. *B.cereus* spores were more resistant to ozonation than the vegetative cells of any of the other organisms tested. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Moat et al 2009[39] |
| | *Bacillus cereus* spores | Ozone | 25ppm ozone for 20 minutes followed by a short burst of humidity >90% resulted in >3.1 $log_{10}$ reduction | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Sharma and Hudson 2008[52] |
| ***Campylobacter jejuni*** | *Campylobacter jejuni* | HINS | Complete inactivation *of C. jejuni* (5 $log_{10}$ CFU/ml reduction) with dose of 18 J/$cm^2$, 30 mins exposure time. *C.jejuni* more sensitive to HINS than *S.enteritidis* and *E.coli* O157:H7. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Murdoch et al 2010[40] |
| ***Clostridium difficile*** | *Clostridium difficile* spores | AM copper AM copper (3 different formulas) | At a concentration of 1ppm, 2/3 formulas (CuAL42 and CuPC33) achieved a 2 $log_{10}$ reduction, while one formula (CuWB50) achieved a 3 $log_{10}$ reduction. All three formulas (on an ultra microfibre cloth) at a concentration of 150ppm removed an initial inoculum of 3x$10^5$ CFU. No CFU were recovered from the cloth after 16 hours. Ultra microfibre cloth and water also removed the | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Gant et al 2007[24] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | inoculum, however CFU were recovered from the cloth after 16 hours. | | |
| *Clostridium difficile* spores | Chlorine dioxide liquid | Chlorine dioxide at a concentration of 630 ± 60 mg/L free chlorine achieved >6 $log_{10}$ reduction (complete inactivation) of spores in Columbia broth in ~10 minutes, however for spores in brain heart infusion broth, ~30 min contact time was required for the same level of activity. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Perez et al 2005[43] |
| *Clostridium difficile* | Chlorine dioxide liquid | A hospital-wide switch from routine cleaning with microfibre and water, and enhanced cleaning with 1,000ppm chlorine releasing agent to cleaning with a commercially available chlorine dioxide product for all routine and terminal cleaning had no significant impact on *C.difficile* infection rates or on *C.difficile* environmental contamination. | SIGN **level 3** evidence McDonald-Arduino **Level V –** Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | Goldenberg et al 2012[7] |
| *Clostridium difficile* spores | Chlorine dioxide liquid (19 different formulations/products) | Only 8/16 products achieved the required reduction in microbial viability (>$10^3$) for contact times of 1 and 60 min, under both clean and dirty conditions. No information was provided on the product concentrations. [The study also included 5 hypochlorite products, none of which achieved adequate disinfection in the exposure time in either clean or dirty conditions.] | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Speight et al 2011[55] |
| *Clostridium difficile* spores and vegetative cells | HINS | Vegetative cells: Significant inactivation was achieved after exposure to a dose of 12 J/$cm^2$, and a 3.7 $log_{10}$ CFU/mL reduction after a dose of 48 J/$cm^2$. *C.difficile* was more readily deactivated than *B.cereus* and *S.aureus*. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction | McLean et al 2013b[36] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | Spores: With exposure to HINS light (405nm) with an irradiance of 40mW/cm$^2$, there was ~1 $log_{10}$ CFU/mL reduction with an exposure to a dose of 0.58kJ/cm$^2$, and a ~2.7 log10 CFU/mL reduction with an exposure of 1.15kJ/cm$^2$. Both reductions were significantly better than the control (no HINS). Significantly greater doses were required for inactivation of spores than vegetative cells. | efficacy | |
| | *Clostridium difficile* HPV | The study demonstrated a reduced risk of *C.difficile* acquisition in patients admitted to rooms decontaminated using HPV compared with rooms decontaminated using standard methods, however this was not significant. [The study looked at a number of multi-drug resistant organisms, and found that HPV decontamination was associated with a 64% lower risk of acquiring an MDRO overall (incidence rate ratio [IRR], 0.36; 95% confidence interval [CI], .19–.70; P < .001).] | SIGN **level 3** evidence McDonald-Arduino **Level V –** Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | Passeretti et al 2013[13] |
| | *Clostridium difficile* spores HPV | Study compared effect of 0.5% hypochlorite versus HPV decontamination of rooms that had accommodated patients with *C.difficile*. Reduction in proportion of contaminated samples was significantly greater for rooms treated with HPV (91%) than hypochlorite (50%) (*P<0* .005). The percentage of rooms with at least 1 sample positive for *C. difficile:* **Hypochlorite disinfection**:<br>Before treatment: 69% (11 of 16)<br>After treatment: 50% (8 of 16) ($\chi^2$=1.17; *P=0*.28). | SIGN **level 3** evidence McDonald-Arduino **Level II –** Demonstration of in-use bioburden reduction effectiveness | Barbut et al 2009[60] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | **HPV disinfection:**<br>Before treatment: 80% (12 of 15)<br>After treatment: 20% (3 of 15) ($\chi^2$=8.53; *P=0*.003).<br>Laboratory study demonstrated a time-dependent reduction: 1.76± 0.96 $\log_{10}$ CFU reduction after 10 seconds; 4.33 ± 0.37 $\log_{10}$ CFU after 20 minutes. | | |
| *Clostridium difficile* | HPV | Before-and-after study comparing standard hypochlorite based terminal cleaning to terminal cleaning followed by HPV. Study demonstrated a 37% reduction in *C.difficile* infection rates (rate ratio = 0.63; 95% confidence interval: 0.50-0.79, P < 0.0001) when HPV was used. | SIGN **level 3** evidence<br>McDonald-Arduino **Level V –** Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | Manian et al 2013[9] |
| *Clostridium difficile* spores and vegetative cells | | All technologies compared to chlorine releasing agent (Actichlor Plus) at 1,000ppm. The three most effective methods in order were HPV, chlorine releasing agent and peracetic acid wipes. | SIGN **level 3** evidence<br>McDonald-Arduino **Level II –** Demonstration of in-use bioburden reduction effectiveness | Doan et al 2012[62] |
| | HPV | 350-700ppm<br>Standardized median CFU $\log_{10}$ reduction (IQR): 2.301 (2.301, 2.301) (P <0.05) | | |
| | Microfibre ± chlorine releasing agent | with 1,000ppm chlorine releasing agent: Standardized median CFU $\log_{10}$ reduction (IQR): 1.523 (0.734, 2.301) (P >0.05)<br>without chlorine releasing agent: Standardized median CFU $\log_{10}$ reduction (IQR): 1.222 | | |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | (0.774, 1.761) (P <0.05) | | |
| | Ozone | 25ppm<br>Standardized median CFU $log_{10}$ reduction (IQR): 1.347 (0.582, 2) (P <0.05) | | |
| | Disinfectant wipes (peracetic acid) | Standardized median CFU $log_{10}$ reduction (IQR): 2.301 (2.151, 2.301) (P >0.05) | | |
| | Steam | Standardized median CFU $log_{10}$ reduction (IQR): 2.0 (1.523, 2.301) (P >0.05) | | |
| | High temperature over heated dry atomized steam | Standardized median CFU $log_{10}$ reduction (IQR): 0.382 (0.017, 0.899) (P <0.05) | | |
| *Clostridium difficile* spores | HPV | Exposure to 1% hydrogen peroxide:<br>• 1 min: inactivation of 75% of spores<br>• 10 min: no change<br>• 20 min: no change<br>Exposure to 10% hydrogen peroxide:<br>• 1 min: inactivation of >99% of spores<br>• 10 min: further inactivation<br>• 20 min: fully inactivated (>6 $log_{10}$ reduction)<br>*G. stearothermophilus* spores ($10^6$ spores) required an exposure to 400 ppm hydrogen peroxide vapour for between 20 and 60 minutes for complete inactivation. In contrast, C.difficile spores required a 5 to 20 minute exposure for complete inactivation. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Lawley et al 2010[28] |
| *Clostridium difficile* spores | Microfibre | Study compared the ability of microfibre cloths and cotton cloths to remove *C.difficile* spores from a ceramic surface, and the ability of cloths to transfer of spores to another surface. Microfibre and cotton cloths removed 2.4 and | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction | Trajtman et al 2015[56] |

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| | | 1.7 $log_{10}$ spores, respectively (initial inoculums ~ 4.2 $log_{10}$). Microfibre and cotton cloths transferred 1.7 and 2.4 $log_{10}$ spores, respectively (initial inoculums ~ 4.2 $log_{10}$). Cloths were pre-wetted with either phosphate buffered saline or $H_2O_2$. Cotton cloths transferred significantly more spores between surfaces than microfibre cloths regardless of whether a detergent was used or not. | efficacy | |
| | *Clostridium difficile* spores | Ozone | Results not clearly reported – 'The maximum log reduction in counts was above the limit of detection (3.20 log) when ozone was used at 25 ppm for 75 min' | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Moat et al 2009[39] |
| | *Clostridium difficile* spores | Ozone | 25ppm ozone for 20 minutes followed by a short burst of humidity >90% resulted in >4.0 $log_{10}$ reduction | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Sharma and Hudson 2008[52] |
| | *Clostridium difficile* spores | Ozone + Hydrogen peroxide | In a test chamber, exposure to 80ppm ozone + 1% hydrogen peroxide at 80% RH resulted in a 7.90 $log_{10}$ reduction after 45, 60 and 90 minutes exposure.<br>In a 113$m^3$ test room, exposure to 80ppm ozone + 1% hydrogen peroxide at 80% RH resulted in a 5.75 $log_{10}$ reduction after 90 minutes exposure. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Zoutman et al 2011[59] |
| | *Clostridium difficile* | Pulsed xenon UV | Hospital before and after study of impact addition of UV disinfection to terminal decontamination on *C.difficile* (and MDRO) | SIGN **level 3** evidence McDonald-Arduino **Level V** – Demonstration of | Haas et al 2014[8] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | rates. *C.difficile* rates decreased from 0.79/1000 patient days before the intervention to 0.65/1000 patient days during the intervention period (Rate ratio 0.83, (95% CI 0.70-0.97), p=0.02).<br>Limitation: simultaneous interventions occurring to reduce acquisition of MDROs and *C.difficile*, so cannot definitively attribute reductions to UV use. | reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | |
| | *Clostridium difficile* | Pulsed xenon UV | Hospital before and after study of impact of addition of UV disinfection to standard manual cleaning of patient areas on *C.difficile* rates. *C.difficile* rates: Baseline period (standard decontamination only) 23.3 per 10,000 patient days; 1st intervention period (introduction of a multidisciplinary *C.difficile* prevention team) 19.3 per 10,000 patient days; 2nd intervention period (UV decontamination + multidisciplinary team) 8.3 per 10,000 patient days. This represents a 56% drop in *C.difficile* rates in the 2nd intervention period compared to the baseline period (p=0.02). The study combines two interventions – so it is not possible to determine whether this impact was solely from the addition of UV or from a synergistic effect of the UV and the multidisciplinary team. | SIGN **level 3** evidence McDonald-Arduino **Level V –** Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | Miller et al 2015[10] |
| | *Clostridium difficile* | Pulsed xenon UV | Hospital before and after study of impact of use of UV decontamination adjunct to traditional cleaning methods on discharge of selected rooms (intensive care unit for all discharges and transfers, but only *C.difficile* rooms for non-ICU discharges and transfers). Intervention resulted | SIGN **level 3** evidence McDonald-Arduino **Level V** – Demonstration of reduced microbial pathogen acquisition (colonisation or infection) | Vianna et al 2016[14] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | in a 41% facility-wide reduction in *C.difficile* infection (p=0.01). There was a 40% reduction in *C.difficile* rates in non-ICU areas (p=0.04). There was a 40% reduction in ICU-rates, but this was not significant (p=0.25).<br>Overall, there was a 29% facility-wide reduction in 3 infections (*C.difficile*, MRSA, VRE) in the intervention period compared to baseline (p=0.01). | by patients via *non-outbreak* surveillance testing and clinical incidence | |
| | *Clostridium difficile* | UV-C | Reduction in *C.difficile* infection incidence rates from 1.23 in the baseline period (terminal cleaning only) to 0.66 in the intervention period (terminal cleaning + UV-C disinfection) (incidence rate change -46.2%, p<0.001). | SIGN **level 3** evidence<br>McDonald-Arduino **Level V** – Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | Napolitano et al 2015[12] |
| | *Clostridium difficile* | Pulsed xenon UV | Hospital study comparing decontamination with activated hydrogen peroxide disinfectant followed by 10% sodium hypochlorite solution, to decontamination with hydrogen peroxide disinfectant followed by pulsed xenon UV disinfection.<br>The mean number of CFUs for the hypochlorite arm decreased by 70% from 2.39 to 0.71 (*p* = 0.14), while the mean number of CFUs for the PX-UV arm decreased significantly by 95% from 22.97 to 1.19 (*p* =0.002). Note: baseline contamination in rooms treated with UV was 8.6 times higher than in the rooms | SIGN **level 2+** evidence<br>McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Ghantoji et al 2015[3] |

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| | | decontaminated with hypochlorite.<br>The difference in final contamination levels between the two cleaning protocols was not significantly significant (P=0.98). | | |
| *Clostridium difficile* spores | Detergent wipes (7 different wipes) | Average removal of C.difficile spores from a stainless steel surface after a 10-second wipe was 0.96 $\log_{10}$ (range 0.26-1.44).<br>Wipes A (Azodet), D (Aquamed), E (Clinitex), and G (Clinell new formulation) removed significantly more spores than wipes B (Clinell old formulation) and C (Sani Cloth).<br>All wipes tested failed to retain spores, with transfer of between 1.29% (Clinell new formulation) and >100% (Sani Cloth).<br>[Wipes performed better for removal and prevention of transfer for *A.baumanii* and *S.aureus* than *C.difficile* spores.] | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Ramm et al 2015[45] |
| *Clostridium difficile* | Disinfectant Wipes (peracetic acid) | Hospital before and after study of impact of changing from chlorine-based cleaning regimen with use of peracetic acid wipes on *C.difficile* infection rates.<br>Mean *C.difficile* infection rate reduced from ~6 per 1,000 patients in the chlorine-based cleaning period to ~2 per 1,000<br>Patients in the period when wipes were used for cleaning. The overall rate of *C.difficile* infection was reduced by 72% following the introduction of the wipes.<br>Limitation: Another intervention was introduced at the same time as wipes (weekly multidisciplinary ward rounds to monitor IPC measures and patient care). It is not possible to | SIGN **level 3** evidence<br>McDonald-Arduino **Level V** – Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | Carter and Barry 2011[5] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | determine what impact the individual interventions had on *C.difficile* infection rates. | | |
| *Clostridium difficile* spores | Disinfectant Wipes (Hydrogen peroxide + peracetic acid; sodium hypochlorite) | The study tested the effectiveness of disinfectants and wipe methods against *C.difficile* spores, particularly the importance of physical removal versus sporicidal inactivation. Study demonstrated that physical removal (i.e. wiping with a non-sporicidal agent) eliminated approximately 3 $\log_{10}$ *C. difficile* spores. Disinfectants with activity against *C. Difficile* spores (e.g. hypochlorite) were highly effective in eliminating spores even without physical removal (> 3 $\log_{10}$ decrease for spray only). Products without activity against *C. difficile* spores were ineffective in eliminating *C. difficile* spores (< 2 $\log_{10}$ decrease) without physical removal. Wiping surfaces twice compared with wiping them once lead to improved removal of *C. Difficile* spores when products without disinfectant activity against *C. difficile* spores were used. Sporicidal disposable wipes were effective in both removing and inactivating the *C. difficile* spores. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Rutala et al 2012[48] |
| *Clostridium difficile* | Disinfectant Wipes (0.55% active chlorine) | Hospital before and after study of impact of changing from use of a quaternary ammonium compound for routine and discharge cleaning to use of chlorine-based wipes. *C.difficile* infection rates reduced by 85%, from 24.2 cases per 10,000 patient days in the pre-intervention period to 3.6 cases per 10,000 patient days in the intervention period ($p<0.001$). | SIGN **level 3** evidence McDonald-Arduino **Level IV** – Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via outbreak surveillance testing and clinical incidence | Orenstein et al 2011[74] |

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| | | The study authors note that the intervention also prolonged the median time between hospital-acquired CDI cases from 8 to 80 days. | | |
| *Clostridium difficile* spores | Disinfectant Wipes (10 different wipes) | The ability of the sporicidal wipes to remove *C. difficile* spores from a surface ranged from 0.22 (1% polymeric biguanide hypochloride, alkyl di-methyl benzyl, ammonium chloride, didecyl dimethyl ammonium chloride) to 4.09 $\log_{10}$ (Wipe A: inorganic peroxygen generator, tetra acetyl ethlenediamine, surfactants) within 10 seconds. One wipe (composition/ingredients unknown) did not remove any spores. None of the wipes demonstrated high sporicidal activity (i.e. 4 $\log_{10}$ reduction) within 5 minutes of contact time, except for a control wipe soaked in 5,000-ppm sodium hypochlorite.<br>Only one wipe (wipe A) demonstrated some sporicidal activity after 5 minutes, with a 1.50 and a 3.74 $\log_{10}$ reduction in spore number of *C.difficile* NCTC12727 and R20291 (ribotype 027), respectively. All but one wipe (wipe A) demonstrated that spores could be repeatedly transferred to other surfaces. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Siani et al 2011[53] |
| *Clostridium difficile* spores | Disinfectant Wipes (4 different wipes) | Study of decontamination of artificially contaminated tablet computers with wipes. *C. difficile* was more difficult to remove than VRE or MRSA. The Tristel Sporicidal wipes (0.1% - 0.12% chlorine dioxide) resulted in the most apparent reduction in *C. difficile* CFU, but this was not significant. None of the wipes tested demonstrated a residual effect on *C. difficile*. | SIGN **level 3** evidence McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Howell et al 2014[66] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| ***Clostridium perfringens*** | *Clostridium perfringens* | HINS | Dose of 45 J/cm$^2$ resulted in 4.4 $\log_{10}$ reduction | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | MacLean et al 2009[35] |
| ***Clostridium sporogenes*** **(*C.difficile* surrogate)** | *Clostridium sporogenes* spores | Disinfectant wipes (5 different wipes) | Only 1/5 wipe performed better than gauze with water at removing spores from the smooth, flat surface of an anaesthetic machine: 0.55% sodium hypochlorite wipe.<br>None of the 5 wipes performed better than gauze and water at removing spores from flat and ridged caps. One wipe performed significantly worse than gauze and water at removing spores from both the flat and ridged caps: 0.28%/17.2%<br>Diisobutylphenoxyethoxyethyl dimethyl benzyl ammonium chloride wipe. Another wipe performed worse than gauze and water at removing spores from the ridged cap: 0.6% citric acid wipe.<br>Authors concluded that wetness of wipes may play a role in effectiveness.<br>[*Clostridium sporogenes* spores were more difficult to clean than *S.aureus*.] | SIGN **level 3** evidence McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Gonzalez et al 2015[65] |
| | *Clostridium sporogenes* spores | Chlorine dioxide liquid | Chlorine dioxide at a concentration of 630 ± 60 mg/L free chlorine achieved >6 $\log_{10}$ reduction (complete inactivation) of spores in Columbia broth in ~10 minutes, however for spores in brain heart infusion broth, ~30 min contact time was required for the same level of activity. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Perez et al 2005[43] |

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| ***Corynebacterium striatum*** | *Corynebacterium striatum* | HINS | Inoculum: 100µL , $10^3$CFU/mL<br>Complete inactivation after 30 mins exposure to 405nm HINS with an average irradiance of 71mW/$cm^2$ | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | McDonald et al 2013[37] |
| ***Escherichia coli*** | *Escherichia coli* | AM silver | Minimum inhibitory concentration of colloidal silver nanoparticles against *E.coli* was found to be 3mg/L (contact time 24 hours).<br>A concentration of 2-3mg/L was found to inhibit growth of *E.coli* in liquid suspension. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Le et al 2012[29] |
| | *Escherichia coli* | AM silver (5 different disinfectants, including 3 different silver nanoparticle solutions) | Agar diffusion tests were used to test activity of 3 different silver nanoparticle solutions: silver nanoparticle solution (NP; 6 mg/ml), concentrated silver nanoparticle solution (CNP; 60 mg/ml), and silver nanoparticle solution containing additional sodium chloride (NPNaCl), and two additional disinfectants dodecyltrimethylammonium bromide (Dotab), silver sulfadiazine (SAg).<br>There was no difference ($P > 0.05$) between the inhibition induced by the NP and NPNaCl treatments, whereas the CNP treatment was the most effective. There was no significant difference ($P > 0.05$) in bacterial inhibition by Dotab and SAg. A smaller inhibition was observed for Dotab and SAg treatments than for the others (NP, CNP, and NPNaCl). | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Araujo et al 2012[15] |
| | *Escherichia coli* | AM silver | The silver nanoparticle solution exhibited full antibacterial activity after 2 hours at 40ppm, whereas phenol and hypochlorite disinfectants | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory | Chamakura et al 2011[20] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | demonstrated full antibacterial activity after 10 minutes at concentrations 16 parts per thousand and 16ppm, respectively.<br>The minimum bactericidal concentration of silver nanoparticle solution was 10 ppm for *S.aureus*, compared to 0.6 ppm for *E.coli* with the same treatment time (4 h). | demonstration of bioburden reduction efficacy | |
| *Escherichia coli* | Chlorine dioxide gas | Complete inactivation at:<br>• 351ppm, 667ppm-hours, 50% RH;<br>• 377ppm, 890ppm-hours, 65% RH;<br>• 379ppm, 767ppm-hour, 65% RH;<br>• 385ppm, 770ppm-hour, 65% RH. | SIGN **level 3** evidence<br>McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Lowe et al 2013c[76] |
| *Escherichia coli* | Electrolysed water | 60 second exposure to slightly acidic electrolysed water (23 mg/L available chlorine) resulted in a 5.07 $\log_{10}$ CFU/mL reduction; 60 second exposure to strongly acidic electrolysed water (50mg/L available chlorine) resulted in a 6.02 $\log_{10}$ CFU/mL reduction; 60 second exposure to NaOCl (120 mg/L available chlorine) resulted in a 5.13 $\log_{10}$ CFU/ml reduction. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Issa-Zacharia et al 2010[27] |
| *Escherichia coli* | Electrolysed water | Pure culture (8.5 log CFU/mL) reduced by more than 7 log CFU/mL after 5 mins exposure to neutral electrolysed water (63mg/L available chlorine) or to NaClO (62 mg/L available chlorine).<br>Stainless steel and glass surfaces inoculated with *E.coli* were rinsed for 1 min in neutral electrolysed water, NaClO or deionised water (control). Both electrolysed water and NaCLO resulted in reductions of more than 6 log CFU/$cm^2$. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Deza et al 2005[22] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| *Escherichia coli* | HINS | Inoculum: 100µL , $10^3$CFU/mL<br>Complete inactivation after 60 mins exposure to 405nm HINS with an average irradiance of 71mW/$cm^2$ | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | McDonald et al 2013[37] |
| *Escherichia coli* O157:H7 | HINS | *E. coli* (on an agar plate) inactivated by 2.18 $log_{10}$ CFU/plate (99.8%), at a dose of 270 J/$cm^2$ and 45 mins exposure time (average irradiance of 71m W/$cm^2$. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Murdoch et al 2012[41] |
| *Escherichia coli* O157:H7 | HINS | *E.coli* O157:H7 inactivated by 5 $log_{10}$ CFU/ml after a total dose of 288 $Jcm^{-2}$.<br>*C.jejuni* more sensitive to HINS than *S.enteritidis* and *E.coli* O157:H7. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Murdoch et al 2010[40] |
| *Escherichia coli* | HINS | Dose of 180 J/$cm^2$ resulted in 3.1 $log_{10}$ reduction | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | MacLean et al 2009[35] |
| *Escherichia coli* | HINS | Negligible inactivation after 30 minutes exposure, however exposures of 45 and 60 min did demonstrate significant differences compared with control samples, indicating that a more prolonged exposure may induce further inactivation. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Maclean et al 2008[34] |
| *Escherichia coli* (biofilm) | HINS | Biofilms grown on glass and acrylic surfaces for 4, 24, 48 and 72 hours and then exposed to | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** | McKenzie et al 2013[38] |

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| | | HINS for 10, 20, 30, 40, 50 and 60 minutes.<br>**Glass**: Fastest inactivation rates for biofilm that was grown for 4 hours, with complete kill after 20 min exposure. After 24 h bacterial biofilm populations were ~ 5.7 $log_{10}$ CFU/mL. Inactivation of these biofilms occurred at a relatively linear rate, with reductions of 2.27, 4.41 and 5.7 $log_{10}$ CFU/mL following exposure to light for 20, 30 and 40 min respectively. Biofilms that had grown for 48-72h had greater cell densities and higher starting populations of 7-8 $log_{10}$ CFU/ml. Inactivation was achieved after 60min exposure.<br>**Acrylic**: Results similar to results for glass. Inactivation occurred at a steady and consistent rate when applied with increasing exposure times of 405 nm light (20, 30, 40 and 60 min), resulting in reductions of 2.30, 3.07, 3.67 and 4.69 $log_{10}$ CFU/mL respectively. Development of biofilms over a 48 h period generated a bacterial population of ~ 5.1 $log_{10}$ CFU/mL, with near-complete inactivation after 60 min exposure. | – Laboratory demonstration of bioburden reduction efficacy | |
| *Escherichia coli* | Ozone | Inoculum: 20µL, 1 x $10^8$ CFU/ml. Exposure to 25ppm ozone for 30 min, with a 10 minute quench to remove residual ozone in a laboratory setting resulted in a 2.02 to 3.87 $log_{10}$ reduction (depending on material – highest log reduction on agar plate, lowest on metal strip).<br>In a test room, *E.coli* was more susceptible to ozonation than the other organisms tested: *C.difficile*, *S.aureus* and *E.faecalis*. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Moat et al 2009[39] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | *Escherichia coli* | Ozone | 25ppm ozone for 20 minutes followed by a short burst of humidity >90% resulted in >3.1 $log_{10}$ reduction | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Sharma and Hudson 2008[52] |
| | *Escherichia coli* | Ozone + hydrogen peroxide | In a test chamber, exposure to 80ppm ozone + 1% hydrogen peroxide at 80% RH resulted in a 6.77 $log_{10}$ reduction after 30, 45, 60 and 90 minutes exposure.<br>In a 113$m^3$ test room, exposure to 80ppm ozone + 1% hydrogen peroxide at 80% RH resulted in a 6.02 $log_{10}$ reduction after 60 minutes exposure. | SIGN **level 3** evidence **Level I** – Laboratory demonstration of bioburden reduction efficacy | Zoutman et al 2011[59] |
| | *Escherichia coli* | Steam – dry steam | 5.0 $log_{10}$ reduction after 3 seconds | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Song et al 2012[54] |
| ***Enterococcus* spp. Including VRE and VSE** | Glycopeptides resistant *Enterococcus* | AM copper (3 different formulas) | 2-3 $log_{10}$ reduction at 1ppm | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Gant et al 2007[24] |
| | *Enterococcus faecalis* | Chlorine dioxide gas | Complete inactivation at:<br>• 351ppm, 667ppm-hours, 50% RH;<br>• 377ppm, 890ppm-hours, 65% RH;<br>• 379ppm, 767ppm-hour, 65% RH;<br>• 385ppm, 770ppm-hour, 65% RH;<br>• 378ppm, 781ppm-hour, 66% RH. | SIGN **level 3** evidence McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Lowe et al 2013c[76] |

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| *Enterococcus faecalis* | HINS | Inoculum: 100µL , $10^3$CFU/mL<br>Complete inactivation after 30 mins exposure to 405nm HINS with an average irradiance of 71mW/cm$^2$ | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | McDonald et al 2013[37] |
| *Enterococcus faecalis* | HINS | Dose of 216 J/cm$^2$ resulted in 2.6 $\log_{10}$ reduction | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | MacLean et al 2009[35] |
| Vancomycin resistant *Enterococci* | HPV | The study demonstrated that patients were 80% less likely to acquire VRE (IRR, 0.20; 95% CI, .08–.52; P < .001) after adjusting for other factors when they were admitted to rooms decontaminated using HPV compared with rooms decontaminated using standard methods.<br>[The study looked at a number of multi-drug resistant organisms, and found that HPV decontamination was associated with a 64% lower risk of acquiring an MDRO overall (incidence rate ratio [IRR], 0.36; 95% confidence interval [CI], .19–.70; P < .001).] | SIGN **level 3** evidence<br>McDonald-Arduino **Level V –** Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | Passeretti et al 2013[13] |
| Vancomycin resistant *Enterococci* | HPV | 15 environmental samples taken from the hospital room of a patient with history of infection and colonisation with VRE, MRSA and gentamycin resistant gram negative rods.<br>Proportion of sites positive for VRE:<br>• Before cleaning: 6.7%<br>• After cleaning(detergent and QAC): 6.7% | SIGN **level 3** evidence<br>McDonald-Arduino **Level III** – Demonstration of in-use bioburden reduction that may be clinically relevant | Otter et al 2007[73] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | • After HPV decontamination: 0<br>No VRE recontamination was identified in the 19 days following use of HPV. | | |
| Vancomycin resistant *Enterococcus faecium* | Microfibre (+ steam) | Before and after hospital (ICU) study comparing routine cleaning with microfibre cloths and water and terminal cleaning with steam and microfibre cloths (intervention) with terminal cleaning with 1,000ppom sodium hypochlorite solution (pre-intervention).<br>VRE incidence reduced after the intervention was introduced, but it increased when cleaning staff hours were reduced following the allocation of additional responsibilities to cleaning staff. A further significant improvement in VRE transmission rates was demonstrated 15 months after introduction of the intervention ($p$ = 0.003). [Limited detail on results presented]. | SIGN **level 3** evidence<br>McDonald-Arduino **Level V** – Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | Gillespie et al 2015[6] |
| *Enterococcus faecalis* | Ozone | Inoculum: 20µL, 1 x $10^8$ CFU/ml. Exposure to 25ppm ozone for 30 min, with a 10 minute quench to remove residual ozone in a laboratory setting resulted in a 2.07 $log_{10}$ reduction on cotton sheet and a 2.79 $log_{10}$ reduction on an agar plate. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Moat et al 2009[39] |
| *Enterococcus faecalis* | Ozone | 25ppm ozone for 20 minutes followed by a short burst of humidity >90% resulted in >3 $log_{10}$ reduction | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Sharma and Hudson 2008[52] |
| Vancomycin resistant | Ozone + hydrogen | In a test chamber, exposure to 80ppm ozone + 1% hydrogen peroxide at 80% RH resulted in a | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** | Zoutman et al 2011[59] |

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| *Enterococcus faecium* | peroxide | 5.79 $log_{10}$ reduction after 30, 45, 60 and 90 minutes exposure.<br>In a 113$m^3$ test room, exposure to 80ppm ozone + 1% hydrogen peroxide at 80% RH resulted in a 6.08 $log_{10}$ reduction after 60 and 90 minutes exposure. | – Laboratory demonstration of bioburden reduction efficacy | |
| High-level aminoglycoside resistant *Enterococcus faecalis* | Steam – dry steam | Kill-time 5 minutes for inoculum with or without bovine serum albumen (initial inoculum $10^9$ CFU/mL). 5 minute kill time for 2.8% sodium hypochlorite with same inoculum concentration in the absence of BSA, but longer kill time required in presence of BSA (i.e. organic soil). | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Bagatinni et al 2015[17] |
| Vancomycin resistant *Enterococcus* | Pulsed xenon UV | Hospital before and after study of impact addition of UV disinfection to terminal decontamination on MDRO (and *C.difficile*) rates. VRE rates decreased from 0.90/1000 patient days before the intervention to 0.73/1000 patient days during the intervention period (Rate ratio 0.82, (95% CI 0.70-0.95), p=0.002).<br>Limitation: simultaneous interventions occurring to reduce acquisition of MDROs and *C.difficile*, so cannot definitively attribute reductions to UV use. | SIGN **level 3** evidence<br>McDonald-Arduino **Level V** – Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | Haas et al 2014[8] |
| Vancomycin resistant *Enterococci* | Pulsed xenon UV | Hospital before and after study of impact on environmental contamination of use of UV decontamination as an adjunct to standard terminal cleaning of isolation rooms that had been occupied by patients with VRE.<br>**Proportion of positive samples:**<br>Before cleaning: 23.3% | SIGN **level 3** evidence<br>McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Stibich et al 2011[72] |

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| | | After cleaning: 8.2%<br>After PX UV treatment: 0<br>**Heterotrophic Plate Counts , CFU/cm$^2$ (mean)**<br>Before cleaning: 33.0<br>After cleaning: 27.4<br>After PX UV treatment: 1.2 | | |
| Vancomycin resistant *Enterococci* | Pulsed xenon UV | Hospital before and after study of impact of use of UV decontamination as an adjunct to traditional cleaning methods on discharge of selected rooms (intensive care unit for all discharges and transfers, but only *C.difficile* rooms for non-ICU discharges and transfers). Intervention resulted in an 87% reduction in VRE rates in ICU areas (p=0.01). There was a non-significant facility-wide reduction in VRE infection (50%, p=0.07). There was a 37% reduction in non-ICU rates, but this was not significant (p=0.27).<br>Overall, there was a 29% facility-wide reduction in 3 infections (*C.difficile*, MRSA, VRE) in the intervention period compared to baseline (p=0.01). | SIGN **level 3** evidence<br>McDonald-Arduino **Level V** – Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | Vianna et al 2016[14] |
| Vancomycin resistant *Enterococci* | UV-C | Non-significant reduction in VRE infection incidence rates from 1 in the baseline period (terminal cleaning only) to 0.88 in the intervention period (terminal cleaning + UV-C disinfection) (incidence rate change -12.30%, p=0.14). | SIGN **level 3** evidence<br>McDonald-Arduino **Level V** – Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | Napolitano et al 2015[12] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| Vancomycin resistant *Enterococci* | Disinfectant wipes (6 different wipes) | Study of efficacy of different wipes against bacteria on artificially contaminated keyboards. Wiping was performed for 5 seconds in a side to side manner on the key surfaces of the entire keyboard, 10 minutes of drying time was allowed.<br>Average CFU decrease after wiping (%):<br>Alcohol wipe: 98.71%<br>CaviWipes (QAC): 100%<br>Chlorine: 99.78%<br>Chlorox disinfecting (QAC): 100%<br>Sani-Cloth Plus (QAC): 100%<br>Vesphene II SE: 100%<br>Sterile water: 99.61%<br>The three wipes containing QAC demonstrated excellent (100%) sustained (48 hours) activity against VRE with an exposure time of 10 minutes. | SIGN **level 3** evidence<br>McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Rutala et al 2006[75] |
| Vancomycin-resistant *Enterococcus faecalis* | Disinfectant wipes (5 different wipes) | Study of decontamination of artificially contaminated plastic surfaces with wipes. Wipes swiped 1, 3 or 5 times over the surface (approximate contact time 1 second per swipe) and allowed to dry for 10 minutes.<br>For all 5 wipe types, swiping the surface 3 or 5 times eliminated more bacteria than only one swipe. Although not statistically significant, a reduction in the number of bacterial colonies was seen with 3 swipes of saline-moistened tissue compared with 1 swipe. At both 3 and 5 swipes for VRE, no type of wipe eliminated significantly more bacteria compared with the others. At one swipe, a chlorhexidine-alcohol wipe, and an accelerated hydrogen peroxide | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Berendt et al 2011[19] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | | wipe eliminated significantly more VRE than the saline-moistened wipe. | | |
| | Vancomycin-resistant *Enterococcus faecium* | Disinfectant Wipes (4 different wipes) | Study of decontamination of artificially contaminated tablet computers with wipes. All wipes were statistically better at removing bacteria from the tablet computer in comparison to the lint free cloth control. Clorox wipes (alkyldimethylbenzyl ammonium chloride 0.184%), Sani-Cloth CHG 2% (70% alcohol and 2% chlorhexidine) and Tristel Sporicidal wipes (0.1% - 0.12% chlorine dioxide) were the most effective. The sani-cloth wipe was the only wipe found to exhibit a residual effect (no growth after recontamination with microbes; up to 12 hours) for VRE (and MRSA). | SIGN **level 3** evidence McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Howell et al 2014[66] |
| ***Francisella tularensis*** | *Francisella tularensis* | Chlorine dioxide gas | Complete activation of spores in 5/6 decontamination trials:<br>• 351ppm, 677ppm-hours, 50% RH;<br>• 377ppm, 890ppm-hours, 65% RH;<br>• 379ppm, 767ppm-hours, 65% RH;<br>• 385ppm, 770ppm-hours, 65% RH;<br>• 376ppm, 788ppm-hours, 64% RH.<br>[Initial inoculum $10^{10}$ CFU] | SIGN **level 3** evidence McDonald-Arduino **Level II –** Demonstration of in-use bioburden reduction effectiveness | Lowe et al 2013a[68] |
| ***Geobacillus stearothermophilus (Bacillus anthracis surrogate)*** | *Geobacillus stearothermophilus* spores | Chlorine dioxide liquid | 1.47 (±0.45) $\log_{10}$ reduction at 500ppm and 3.07 (±0.09) $\log_{10}$ reduction at 1000ppm after 5 minutes contact time at 22 ± 2°C. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Sabbah et al 2010[50] |
| | *Geobacillus stearothermophilus* spores | HPV | *G. stearothermophilus* spores ($10^6$ spores) required an exposure to 400 ppm hydrogen peroxide vapour for between 20 and 60 | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory | Lawley et al 2010[28] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | | minutes for complete inactivation. In contrast, C.difficile spores required a 5 to 20 minute exposure for complete inactivation. | demonstration of bioburden reduction efficacy | |
| ***Haemophilus influenzae*** | *Haemophilus influenzae* | Ozone | 25ppm ozone for 20 minutes followed by a short burst of humidity >90% resulted in ≥4 $log_{10}$ reduction | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Sharma and Hudson 2008[52] |
| ***Klebsiella* spp.** | *Klebsiella pneumoniae* | HINS | Inoculum: 100µL , $10^3$CFU/mL Complete inactivation after 40 mins exposure to 405nm HINS with an average irradiance of 71mW/$cm^2$ | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | McDonald et al 2013[37] |
| | *Klebsiella pneumoniae* | HINS | Dose of 180 J/$cm^2$ resulted in 3.9 $log_{10}$ reduction | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | MacLean et al 2009[35] |
| | *Klebsiella pneumoniae* | Ozone | 25ppm ozone for 20 minutes followed by a short burst of humidity >90% resulted in ≥4 $log_{10}$ reduction | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Sharma and Hudson 2008[52] |
| | Carbapenemase-producing *Klebsiella pneumoniae* | Steam – dry steam | Kill-time 5 minutes for inoculum with or without bovine serum albumen (initial inoculum $10^9$ CFU/mL). 5 minute kill time for 2.8% sodium hypochlorite with same inoculum concentration in the absence of BSA, but longer | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction | Bagatinni et al 2015[17] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | | kill time required in presence of BSA (i.e. organic soil). | efficacy | |
| | *Klebsiella pneumoniae* | UV-C | Reduction in *Klebsiella pneumoniae* infection incidence rates from 0.44 in the baseline period (terminal cleaning only) to 0.00 in the intervention period (terminal cleaning + UV-C disinfection) (incidence rate change -100%, p<0.001). | SIGN **level 3** evidence McDonald-Arduino **Level V** – Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | Napolitano et al 2015[12] |
| ***Legionella pneumophila*** | *Legionella pneumophila* | AM copper (3 different formulas) | At a concentration of 1ppm, 2/3 formulas (CuAL42 and CuWB50) achieved a 2 $\log_{10}$ reduction and one formula (CuPC33) achieved a 3 $\log_{10}$ reduction. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Gant et al 2007[24] |
| | *Legionella pneumophila* | Ozone | 25ppm ozone for 20 minutes followed by a short burst of humidity >90% resulted in ≥4 $\log_{10}$ reduction | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Sharma and Hudson 2008[52] |
| ***Listeria* spp.** | *Listeria monocytogenes* | Electrolysed water | Pure culture (8.5 log CFU/mL) reduced by more than 7 log CFU/mL after 5 mins exposure to neutral electrolysed water (63mg/L available chlorine) or to NaClO (62 mg/L available chlorine).<br>Stainless steel and glass surfaces inoculated with *Listeria monocytogenes* were rinsed for 1 min in neutral electrolysed water, NaClO or | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Deza et al 2005[22] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | deionised water (control). Both electrolysed water and NaCLO resulted in reductions of more than 6 log CFU/$cm^2$. | | |
| | *Listeria innocua* | AM silver (5 different disinfectants, including 3 different silver nanoparticle solutions) | Agar diffusion tests were used to test activity of 3 different silver nanoparticle solutions: silver nanoparticle solution (NP; 6 mg/ml), concentrated silver nanoparticle solution (CNP; 60 mg/ml), and silver nanoparticle solution containing additional sodium chloride (NPNaCl) and two additional disinfectants dodecyltrimethylammonium bromide (Dotab), silver sulfadiazine (SAg). In the case of *L. innocua*, the antimicrobial action was the same for the NP, CNP, and NPNaCl treatments. There was no difference ($P > 0.05$) between the inhibition induced by the NP and NPNaCl treatments, whereas the CNP treatment was the most effective. There was no significant difference ($P > 0.05$) in bacterial inhibition by Dotab and Sag. A smaller inhibition was observed for Dotab and SAg treatments than for the others (NP, CNP, and NPNaCl). | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Araujo et al 2012[15] |
| | *Listeria monocytogenes* | HINS | *L.monocytogenes* (on an agar plate) inactivated by 2.25 $log_{10}$ CFU/plate (100%), at a dose of 180 J/$cm^2$ and 30 mins exposure time (average irradiance of 71m W/$cm^2$). *L.monocytogenes* (on PVC) 0.90 $log_{10}$ CFU (90%) inactivation at a dose of 45 J/$cm^2$ and 7.5 mins exposure time (irradiance of 110 W/$cm^2$). *L.monocytogenes* (on acrylic) 0.42 $log_{10}$ CFU (61%) inactivation at a dose of 60 J/$cm^2$ and 10 mins exposure time (irradiance of 110 W/$cm^2$). | SIGN **level 3** evidence **Level I** – Laboratory demonstration of bioburden reduction efficacy | Murdoch et al 2012[41] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| *Listeria monocytogenes* (biofilm) | HINS | Biofilms on glass exposed to HINs for 5, 10 and 20 minutes.<br>5 mins exposure (42 J/cm$^2$) resulted in a 0.61 $\log_{10}$ CFU/mL reduction. 10 mins exposure (84 J/cm$^2$) resulted in a 1.09 $\log_{10}$ CFU/mL reduction. 20 mins exposure (168 J/cm$^2$) resulted in a 2.48 $\log_{10}$ CFU/mL reduction. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | McKenzie et al 2013[38] |
| *Listeria monocytogenes* | Ozone | Inoculum: 20µL, 1 x $10^8$ CFU/ml. Exposure to 25ppm ozone for 30 min, with a 10 minute quench to remove residual ozone in a laboratory setting resulted in a 2.19 to 3.38 $\log_{10}$ reduction (depending on material – highest log reduction on cotton sheet, lowest on Formica laminate). | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Moat et al 2009[39] |
| *Listeria monocytogenes* (surface attached bacteria and biofilms) | Ozone/Open air factor (OAF) | Exposure to ozone concentrations of 2, 5 and 10 ppm for 1 hour resulted in log reductions in 0.24, 0.33 and 0.57, respectively, of cells surface attached on stainless steel.<br>Ozone concentrations of 45 ppm resulted in 3.41 and 3.42 $\log_{10}$ reductions, on stainless steel and granite respectively, compared with polypropylene which gave a 1.11 $\log_{10}$ reduction. Open air factor reduced the number of surface attached *L. monocytogenes* on stainless steel by 1.86 $\log_{10}$. OAF gave better log reductions in surface attached cells than 10 ppm ozone, but lower log reductions than 45 ppm. Conversely, OAF was significantly better than ozone at reducing the number of biofilm organisms. Biofilm organisms were significantly more resistant than surface-attached cells on stainless steel. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Nicholas et al 2013[42] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | *Listeria monocytogenes* (biofilms) | Ozone (± sonication) | The greatest reduction was observed with exposure to 1.0 ppm ozone for 60s (4.2-log CFU/ml reduction).<br>There was a significant difference between the results of sonication (20kHz, 100% amplitude, 120 W) and ozonation when they were used separately. The reduction of cell numbers due to sonication was greater at both 30 and 60 s than any of the ozone concentrations or times used. The simultaneous use of low ozone concentrations with sonication appeared to have additive effects.<br>Higher concentrations of ozone resulted in a synergistic effect when used with sonication for 60 s. This was not observed at 30s. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Baumann et al 2009[18] |
| ***Micrococcus* spp.** | *Micrococcus spp.* | HINS | Inoculum: 100µL , $10^3$CFU/mL<br>Complete inactivation after 20 mins exposure to 405nm HINS with an average irradiance of 71mW/$cm^2$ | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | McDonald et al 2013[37] |
| ***Mycobacterium smegmatis*** | *Mycobacterium smegmatis* | Chlorine dioxide gas | Complete inactivation in ¾ decontamination trials:<br>• 362ppm, 850ppm-hours, 55% RH;<br>• 406ppm, 763ppm-hour, 65% RH;<br>• 695ppm, 756ppm hours, 55% RH.<br>[Initial inoculum $10^{10}$ CFU] | SIGN **level 3** evidence<br>McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Lowe et al 2013b[67] |
| | *Mycobacterium smegmatis* | Chlorine dioxide gas | Complete inactivation at:<br>• 351ppm, 667ppm-hours, 50% RH;<br>• 377ppm, 890ppm-hours, 65% RH;<br>• 379ppm, 767ppm-hour, 65% RH;<br>• 385ppm, 770ppm-hour, 65% RH; | SIGN **level 3** evidence<br>McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Lowe et al 2013c[76] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | *Mycobacterium smegmatis* | HINS | At least a 100 J/cm$^2$ dose was required for effective inactivation.<br>Doses of 120J/cm$^2$ (98.3% kill rate), 150J/cm$^2$ (96.7% kill rate), and 215J/cm$^2$ (100% kill rate) were the most effective at inactivating *M.smegmatis* ($P \leq 0.001$).<br>The dose response relationship was not linear. Some degree of effectiveness was lost at 180J/cm$^2$ and 250J/cm$^2$. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Guffey et al 2013[25] |
| | *Mycobacterium smegmatis* | Ozone | 25ppm ozone for 20 minutes followed by a short burst of humidity >90% resulted in >2.7 $\log_{10}$ reduction | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Sharma and Hudson 2008[52] |
| ***Mycobacterium terrae*** | *Mycobacterium terrae* | Chlorine dioxide liquid | >8 $\log_{10}$ reduction after 5 minutes contact time at 22 ± 2°C for both 500ppm and 1000ppm. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Sabbah et al 2010[50] |
| ***Proteus vulgaris*** | *Proteus vulgaris* | HINS | Dose of 144 J/cm$^2$ resulted in 4.7 $\log_{10}$ reduction | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | MacLean et al 2009[35] |
| ***Pseudomonas aeruginosa*** | *Pseudomonas aeruginosa* | Electrolysed water | Pure culture (8.5 log CFU/mL) reduced by more than 7 log CFU/mL after 5 mins exposure to neutral electrolysed water (63mg/L available chlorine) or to NaClO (62 mg/L available chlorine). | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction | Deza et al 2005[22] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | Stainless steel and glass surface inoculated with *P.aeruginosa* were rinsed for 1 min in neutral electrolysed water, NaClO or deionised water (control). Both electrolysed water and NaCLO resulted in reductions of more than 6 log CFU/$cm^2$. | efficacy | |
| *Pseudomonas aeruginosa* | AM silver (5 different disinfectants, including 3 different silver nanoparticle solutions) | Agar diffusion tests were used to test activity of 3 different silver nanoparticle solutions: silver nanoparticle solution (NP; 6 mg/ml), concentrated silver nanoparticle solution (CNP; 60 mg/ml), and silver nanoparticle solution containing additional sodium chloride (NPNaCl) and two additional disinfectants dodecyltrimethylammonium bromide (Dotab), silver sulfadiazine (SAg). There was no difference ($P > 0.05$) between the inhibition induced by the NP and NPNaCl treatments, whereas the CNP treatment was the most effective. There was no significant difference ($P > 0.05$) in bacterial inhibition by Dotab and SAg. A smaller inhibition was observed for Dotab and SAg treatments than for the others (NP, CNP, and NPNaCl).<br>The study also demonstrated that the CNP treatment was the most effective for removal/destruction of *P.aeruginosa* cells adhered to a stainless steel surface, producing a 5-log reduction of the microbial population after 30 to 60 min of immersion. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Araujo et al 2012[15] |
| *Pseudomonas aeruginosa* | HINS | Inoculum: 100µL , $10^3$CFU/mL<br>Complete inactivation after 12 mins exposure to 405nm HINS with an average irradiance of | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory | McDonald et al 2013[37] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | 71mW/cm$^2$ | demonstration of bioburden reduction efficacy | |
| *Pseudomonas aeruginosa* | HINS | Dose of 180 J/cm$^2$ resulted in 4.2 $\log_{10}$ reduction | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | MacLean et al 2009[35] |
| *Pseudomonas aeruginosa* (biofilm) | HINS | Biofilms on glass exposed to HINs for 5, 10 and 20 minutes. 5 mins exposure (42 J/cm$^2$) resulted in a 1.5 $\log_{10}$ CFU/mL reduction. 10 mins exposure (84 J/cm$^2$) resulted in a 2.43 $\log_{10}$ CFU/mL reduction. 20 mins exposure (168 J/cm$^2$) resulted in a 3.72 $\log_{10}$ CFU/mL reduction. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | McKenzie et al 2013[38] |
| *Pseudomonas aeruginosa* (resistant to ceftazidime or imipenem) | HPV | All *P.aeruginosa* contamination in all rooms tested removed by terminal cleaning alone, so HPV not necessary for removal of this organism. [Only 1/1456 of samples were contaminated with *P.aeruginosa* after patient discharge, so there was a low level of initial contamination.] | SIGN **level 3** evidence McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Blazejewski et al 2015[61] |
| *Pseudomonas aeruginosa* | Ozone | 25ppm ozone for 20 minutes followed by a short burst of humidity >90% resulted in ≥4 $\log_{10}$ reduction | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Sharma and Hudson 2008[52] |
| *Pseudomonas aeruginosa* | Ozone + hydrogen peroxide | In a test chamber, exposure to 80ppm ozone + 1% hydrogen peroxide at 80% RH resulted in a 7.36 $\log_{10}$ reduction after 30, 45, 60 and 90 | SIGN **level 3** evidence **Level I** – Laboratory demonstration of | Zoutman et al 2011[59] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | | minutes exposure. | bioburden reduction efficacy | |
| | *Pseudomonas aeruginosa* | Steam – dry steam | 5.3 $log_{10}$ reduction after 5 seconds | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Song et al 2012[54] |
| | *Pseudomonas aeruginosa* | Steam – dry steam | Kill-time 5 minutes for inoculum with or without bovine serum albumen (initial inoculum $10^9$ CFU/mL). 5 minute kill time for 2.8% sodium hypochlorite with same inoculum concentration in the absence of BSA, but longer kill time required in presence of BSA (i.e. organic soil). | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Bagatinni et al 2015[17] |
| | *Pseudomonas aeruginosa* | Disinfectant wipes (5 different wipes) | Study of efficacy of different wipes against bacteria on artificially contaminated keyboards. Wiping was performed for 5 seconds in a side to side manner on the key surfaces of the entire keyboard, 10 minutes of drying time was allowed.<br>Average CFU decrease after wiping (%):<br>Alcohol wipe: 98.99%<br>CaviWipes (QAC): 99.69%<br>Chlorine: 100%<br>Chlorox disinfecting (QAC): 99.89%<br>Sani-Cloth Plus (QAC): 100%<br>Vesphene II SE: 100%<br>Sterile water: 99.75% | SIGN **level 3** evidence McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Rutala et al 2006[75] |
| ***Propionibacterium acnes*** | *Propionibacterium acnes* | Ozone | 25ppm ozone for 20 minutes followed by a short burst of humidity >90% resulted in ≥4.0 $log_{10}$ reduction | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory | Sharma and Hudson 2008[52] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | | | demonstration of bioburden reduction efficacy | |
| ***Salmonella* spp.** | *Salmonella* spp. | Electrolysed water | 60 second exposure to slightly acidic electrolysed water (23 mg/L available chlorine) resulted in a 5.18 $log_{10}$ CFU/mL reduction; 60 second exposure to strongly acidic electrolysed water (50mg/L available chlorine) resulted in a 6.12 $log_{10}$ CFU/mL reduction; 60 second exposure to NaOCl (120 mg/L available chlorine) resulted in a 5.22 $log_{10}$ CFU/ml reduction. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Issa-Zacharia et al 2010[27] |
| ***Salmonella choleraesuis*** | *Salmonella choleraesuis* | AM silver (5 different disinfectants, including 3 different silver nanoparticle solutions) | Agar diffusion tests were used to test activity of 3 different silver nanoparticle solutions: silver nanoparticle solution (NP; 6 mg/ml), concentrated silver nanoparticle solution (CNP; 60 mg/ml), and silver nanoparticle solution containing additional sodium chloride (NPNaCl) and two additional disinfectants dodecyltrimethylammonium bromide (Dotab), silver sulfadiazine (SAg). There was no difference ($P > 0.05$) between the inhibition induced by the NP and NPNaCl treatments, whereas the CNP treatment was the most effective. There was no significant difference ($P > 0.05$) in bacterial inhibition by Dotab and SAg. A smaller inhibition was observed for Dotab and SAg treatments than for the others (NP, CNP, and NPNaCl). | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Araujo et al 2012[15] |
| ***Salmonella enterica*** | *Salmonella enterica* | HINS | *S.enterica* (on an agar plate) inactivated by 2.28 $log_{10}$ CFU/plate (100%), at a dose of 270 J/$cm^2$ and 45 mins exposure time (average irradiance | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory | Murdoch et al 2012[41] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | | of 71m W/cm$^2$.<br>*S.enterica* (on PVC) 2.19 log$_{10}$ CFU (100%) inactivation at a dose of 45 J/cm$^2$ and 7.5 mins exposure time (irradiance of 110 W/cm$^2$).<br>*S.enterica* (on acrylic) 1.63 log$_{10}$ CFU (98%) inactivation at a dose of 60 J/cm$^2$ and 10 mins exposure time (irradiance of 110 W/cm$^2$). | demonstration of bioburden reduction efficacy | |
| ***Salmonella enteritidis*** | *Salmonella enteritidis* | HINS | *E.coli* O157:H7 inactivated by 3 log$_{10}$ CFU/ml after a total dose of 288 Jcm$^{-2}$.<br>*C.jejuni* more sensitive to HINS than *S.enteritidis* and *E.coli* O157:H7. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Murdoch et al 2010[40] |
| ***Serratia marcescens*** | *Serratia marcescens* | HINS | Inoculum: 100µL , 10$^3$CFU/mL<br>Complete inactivation after 60 mins exposure to 405nm HINS with an average irradiance of 71mW/cm$^2$ | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | McDonald et al 2013[37] |
| ***Shigella* spp.** | *Shigella sonnei* | HINS | *S.sonnei* (on an agar plate) inactivated by 2.10 log$_{10}$ CFU/plate (99.3%), at a dose of 270 J/cm$^2$ and 45 mins exposure time (average irradiance of 71m W/cm$^2$. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Murdoch et al 2012[41] |
| ***Staphylococcus spp.*** | *Staphylococcus spp.* | HINS | ICU-based study.<br>Study 1: Occupied isolation room. Mean CFU/plate 29.0 (SE ± 2.9) before exposure to HINS. Mean CFU/plate 9.6 (SE ± 2.9) after 5-days exposure to HINS. This represented a 66.8% reduction (p=0.0001).<br>Study 2: Occupied isolation room. Mean | SIGN **level 3** evidence<br>McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | MacLean et al 2013a[70] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | | CFU/plate 22.4 (SE ± 6.0) before exposure to HINS. Mean CFU/plate 13.9 (SE ± 6.0) after 1-day exposure to HINS. This represented a non-significant 38% reduction (p=0.42). Mean CFU/plate 63.4 (SE ± 6.0) after HINS had been switched off for 24 hours. This represented a significant 375% increase (p=0.000). | | |
| ***Staphylococcus aureus*, including MRSA** | Meticillin resistant *Staphylococcus aureus* | AM copper (3 different formulas) | At a concentration of 1ppm, all three formulations achieved a 2 to 3 $log_{10}$ reduction (CuAL42, CuPC33 and CuWB50).<br>All three formulas (on an ultra microfibre cloth) at a concentration of 150ppm removed an initial inoculum of $2x10^6$ CFU. No CFU were recovered from the cloth after 16 hours. Ultra microfibre cloth and water also removed the inoculum, however CFU were recovered from the cloth after 16 hours. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Gant et al 2007[24] |
| | Meticillin resistant *Staphylococcus aureus* (169 different isolates) | AM Copper (2 different formulas) | The growth of all 169 MRSA isolates was inhibited by CuAL42, with both the minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) ≤18.75 ppm. One isolate had an MBC of 37.5 ppm with CuWB50.<br>The time-kill curve studies demonstrated that 150 ppm of CuAL42 and CuWB50 produced a 6 $log_{10}$ reduction in bacterial numbers in 1 and 1.5 h, respectively, which was reduced to 30 and 45 min, respectively, at 300ppm. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Luna et al 2010[33] |
| | *Staphylococcus aureus* | AM silver (5 different disinfectants, including 3 | Agar diffusion tests were used to test activity of 3 different silver nanoparticle solutions: silver nanoparticle solution (NP; 6 mg/ml), concentrated silver nanoparticle solution (CNP; | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of | Araujo et al 2012[15] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | different silver nanoparticle solutions) | 60 mg/ml), and silver nanoparticle solution containing additional sodium chloride (NPNaCl) and two additional disinfectants dodecyltrimethylammonium bromide (Dotab), silver sulfadiazine (SAg). There was no difference ($P > 0.05$) between the inhibition induced by the NP and NPNaCl treatments, whereas the CNP treatment was the most effective. | bioburden reduction efficacy | |
| *Staphylococcus aureus* | AM silver | The minimum bactericidal concentration of silver nanoparticle solution was 10 ppm for *S.aureus*, compared to 0.6 ppm for *E.coli* with the same treatment time (4 h). | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Chamakura et al 2011[20] |
| *Staphylococcus aureus* | Chlorine dioxide gas | Incomplete inactivation in all 4 decontamination trials:<br>• 92% inactivation: 362ppm; 850ppm-hours; 55% RH<br>• 95% inactivation: 315ppm; 479ppm-hours; 65% RH<br>• 99% inactivation: 406ppm; 763ppm-hours; 65% RH<br>• 97% inactivation: 695ppm; 756ppm-hours; 55% RH<br>[Initial inoculum $10^{10}$ CFU] | SIGN **level 3** evidence McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Lowe et al 2013b[67] |
| *Staphylococcus aureus* | Chlorine dioxide gas | Complete inactivation at:<br>• 351ppm, 667ppm-hours, 50% RH;<br>• 377ppm, 890ppm-hours, 65% RH;<br>• 379ppm, 767ppm-hour, 65% RH;<br>• 385ppm, 770ppm-hour, 65% RH; | SIGN **level 3** evidence McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Lowe et al 2013c[76] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| *Staphylococcus aureus* | Electrolysed water | 60 second exposure to slightly acidic electrolysed water (23 mg/L available chlorine) resulted in a 4.83 $log_{10}$ CFU/mL reduction; 60 second exposure to strongly acidic electrolysed water (50mg/L available chlorine) resulted in a 4.91 $log_{10}$ CFU/mL reduction; 60 second exposure to NaOCl (120 mg/L available chlorine) resulted in a 5.22 $log_{10}$ CFU/ml reduction. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Issa-Zacharia et al 2010[27] |
| *Staphylococcus aureus* | Electrolysed water | Pure culture (8.5 log CFU/mL) reduced by more than 7 log CFU/mL after 5 mins exposure to neutral electrolysed water (63mg/L available chlorine) or to NaClO (62 mg/L available chlorine).<br>Stainless steel and glass surface inoculated with *Staphylococcus aureus* were rinsed for 1 min in neutral electrolysed water, NaClO or deionised water (control). Both electrolysed water and NaCLO resulted in reductions of more than 6 log CFU/$cm^2$. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Deza et al 2005[22] |
| *Staphylococcus aureus* | HINS | Inoculum: 100µL , $10^3$CFU/mL<br>Complete inactivation after ~13mins exposure to 405nm HINS with an average irradiance of 71mW/$cm^2$<br>Exposure to 5mW/cm2 for 1 hour reduced bacterial contamination by 98%. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | McDonald et al 2013[37] |
| *Staphylococcus aureus* (presumptive) | HINS | Study conducted in a burns unit.<br>Study A: unoccupied room. Mean CFU count 7.02 (SE ±0.66) before exposure to HINS. Mean CFU count 0.56 (SE ±0.66) after 24 hours exposure to HINS. This represents a significant 92% reduction. Mean CFU count 0.89 (SE ±0.68) | SIGN **level 3** evidence McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Maclean et al 2010[69] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | after HINS had been switched off for 24 hours. This represents a 60% increase (not significant).<br>Study B1: Room occupied by an MRSA colonised patient. Mean CFU count 8.52 (SE ±0.59) before exposure to HINS. Mean CFU count 2.94 (SE ±0.59) after 2 days exposure to HINS. This represents a significant 65% reduction.<br>Study B2: Room occupied by an MRSA colonised patient. Mean CFU count 8.52 (SE ±0.59) before exposure to HINS. Mean CFU count 2.08 (SE ±0.59) after 5 days exposure to HINS. This represents a significant 76% reduction.<br>Study C: Room occupied by an MRSA colonised patient. Mean CFU count 36.74 (SE ±6.17) before exposure to HINS. Mean CFU count 18.38 (SE ±6.19) after 5 days exposure to HINS. This represents a significant 50% reduction. Mean CFU count 36.34 (SE ±6.17) after HINS had been switched off for 6 days. This represents a 98% increase (significant). | | |
| Meticillin resistant *Staphylococcus aureus and* meticiliin s*usceptible Staphylococcus aureus* | HINS | MSSA: 5 $\log_{10}$ reduction with dose of 630J/cm (350mW/cm$^2$ x 30 min). Longer exposure times required for same reduction in MRSA (~60 mins).<br>The maximum $\log_{10}$ reduction of *S. aureus* cells resulted from exposure to 405±5 nm wavelength light. Exposure to bandwidths of 430–500nm did not cause significant inactivation of the bacteria. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Maclean et al 2008[34] |

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| *Meticillin resistant Staphylococcus aureus and meticiliin susceptible Staphylococcus aureus* | HINS | MSSA: Dose of 36 J/cm$^2$ resulted in 5 $\log_{10}$ reduction<br>MRSA: Dose of 45 J/cm$^2$ resulted in 5 $\log_{10}$ reduction | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | MacLean et al 2009[35] |
| *Staphylococcus aureus* | HINS | Significant inactivation was observed after exposure to 28.8 J/cm$^2$, with no detectable survival (<1 CFU/mL) after a dose of 72 J/cm$^2$. *S.aureus* was more susceptible to inactivation than *B.cereus* vegetative cells, but less susceptible than *C.difficile* vegetative cells. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | McLean et al 2013b[36] |
| *Staphylococcus aureus* (biofilm) | HINS | Biofilms on glass exposed to HINs for 5, 10 and 20 minutes.<br>5 mins exposure (42 J/cm$^2$) resulted in a 0.6 $\log_{10}$ CFU/mL reduction. 10 mins exposure (84 J/cm$^2$) resulted in a 1.87 $\log_{10}$ CFU/mL reduction. 20 mins exposure (168 J/cm$^2$) resulted in a 2.75 $\log_{10}$ CFU/mL reduction. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | McKenzie et al 2013[38] |
| Meticillin resistant *Staphylococcus aureus* | HPV | The study demonstrated a reduced risk of MRSA acquisition in patients admitted to rooms decontaminated using HPV compared with rooms decontaminated using standard methods, however this was not significant. [The study looked at a number of multi-drug resistant organisms, and found that HPV decontamination was associated with a 64% lower risk of acquiring an MDRO overall (incidence rate ratio [IRR], 0.36; 95% confidence interval [CI], .19–.70; P < .001).] | SIGN **level 3** evidence<br>McDonald-Arduino **Level V –** Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | Passeretti et al 2013[13] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| Meticillin resistant *Staphylococcus aureus* | HPV | Hospital-based study comparing standard terminal cleaning to HPV decontamination of rooms that had recently accommodated MRSA patients. Typical HPV exposure 500ppm for 40 minutes.<br>**Rooms subject to terminal cleaning (detergent only):** 90% of swabs MRSA positive before cleaning and 66% positive after cleaning (p value not reported).<br>**Rooms subject to HPV decontamination:** 72% of swabs were positive for MRSA before decontamination, and 1.2% of swabs were positive after HPV decontamination (p value not reported). | SIGN **level 3** evidence<br>McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | French et al 2004[63] |
| Meticillin resistant *Staphylococcus aureus* | HPV | 15 environmental samples taken from the hospital room of a patient with history of infection and colonisation with VRE, MRSA and gentamycin resistant gram negative rods.<br>Proportion of sites positive for MRSA:<br>• Before cleaning: 60%<br>• After cleaning(detergent and QAC): 40%<br>• After HPV decontamination: 3.3%<br>MRSA recontamination was identified 5 days after HPV decontamination. | SIGN **level 3** evidence<br>McDonald-Arduino **Level III** – Demonstration of in-use bioburden reduction that may be clinically relevant | Otter et al 2007[73] |
| Meticillin resistant *Staphylococcus aureus* | HPV | Hospital-based before and after study comparing the impact of detergent-only terminal cleaning with HPV decontamination of rooms that had been occupied by MRSA colonised/infected patients.<br>MRSA was isolated from 24.7% rooms following detergent cleaning and from 18.8% of rooms after HPV decontamination (p<0.001). The | SIGN **level 3** evidence<br>McDonald-Arduino **Level V** – Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance | Mitchell et al 2014[11] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | incidence of MRSA acquisition reduced from 9.0 to 5.3 per 10 000 patient days in detergent and disinfectant arms, respectively ($p<0.001$). [The study used detergent only for terminal cleaning – this is not routine practice in Scotland. In addition, MRSA screening and monitoring was introduced concurrently with HPV decontamination, so it is not possible to attribute the reduction in incidence to HPV.] | testing and clinical incidence | |
| Meticillin resistant *Staphylococcus aureus* | HPV | All MRSA contamination in all rooms tested removed by terminal cleaning alone, so HPV not necessary for removal of this organism. [Only 0.27% of samples were contaminated with MRSA after patient discharge, so there was a low level of initial contamination.] | SIGN **level 3** evidence McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Blazejewski et al 2015[61] |
| *Staphylococcus aureus* | Ozone | Inoculum: 20µL, 1 x $10^8$ CFU/ml. Exposure to 25ppm ozone for 30 min, with a 10 minute quench to remove residual ozone in a laboratory setting resulted in a 2.33 to 3.30 $log_{10}$ reduction (depending on material – highest log reduction on cotton sheet, lowest on Formica laminate). | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Moat et al 2009[39] |
| *Staphylococcus aureus* (meticillin susceptible and meticillin resistant) | Ozone | MRSA: 25ppm ozone for 20 minutes followed by a short burst of humidity >90% resulted in >3.0 $log_{10}$ reduction<br>MSSA: 25ppm ozone for 20 minutes followed by a short burst of humidity >90% resulted in >2.5 $log_{10}$ reduction | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Sharma and Hudson 2008[52] |
| Meticillin resistant *Staphylococcus aureus* | Ozone/ozone + hydrogen peroxide | MRSA was exposed to ozone (without hydrogen peroxide) in increasing concentrations at 80% humidity for 90 minutes. At 50-180 ppm ozone, bacterial kill was negligible; however, at 500 | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of | Zoutman et al 2011[59] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | ppm ozone, there was a >6 $log_{10}$ reduction in bacteria compared with the unexposed control discs.<br>The bacterial kill was enhanced by increasing the humidity level to 80% compared with 45% or 60% humidity. A >7 $log_{10}$ reduction was achieved after 90 minutes of exposure to 80ppm ozone and 0.2% hydrogen peroxide at 80% humidity.<br>Under these same conditions of ozone and humidity but without any hydrogen peroxide, the bacterial kill was minimal, suggesting that hydrogen peroxide was producing a synergistic bacterial kill with the ozone gas compared with the same conditions without hydrogen peroxide. | bioburden reduction efficacy | |
| *Staphylococcus aureus* | Steam – dry steam | 5.3 $log_{10}$ reduction after 5 seconds | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Song et al 2012[54] |
| Meticillin resistant *Staphylococcus aureus* | Steam – dry steam | Kill-time 5 minutes for inoculum with or without bovine serum albumen (initial inoculum $10^9$ CFU/mL). 5 minute kill time for 2.8% sodium hypochlorite with same inoculum concentration in the absence of BSA, but longer kill time required in presence of BSA (i.e. organic soil). | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Bagatinni et al 2015[17] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| Meticillin resistant *Staphylococcus aureus* | Pulsed xenon UV | Hospital before and after study of impact addition of UV disinfection to terminal decontamination on MDRO (and *C.difficile*) rates. MRSA rates decreased from 0.45/1000 patient days before the intervention to 0.33/1000 patient days during the intervention period (Rate ratio 0.73, (95% CI 0.58-0.92), p=0.007).<br>Limitation: simultaneous interventions occurring to reduce acquisition of MDROs and *C.difficile*, so cannot definitively attribute reductions to UV use. | SIGN **level 3** evidence<br>McDonald-Arduino **Level V** – Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | Haas et al 2014[8] |
| Meticillin resistant *Staphylococcus aureus* | Pulsed xenon UV | Hospital study comparing the effect of manual terminal cleaning (with a hypochlorite solution) versus UV disinfection on MRSA contamination of high-touch surfaces in rooms vacated by patients with MRSA. 20 rooms in each arm of the study. Outcome measure; mean colony count, median (IQR):<br>Before manual cleaning: 127.3; 28.5 (18-143)<br>After manual cleaning: 11.3; 1.0 (0-4) (91% reduction)<br>Before UV disinfection: 108.2; 123.0 (14-183)<br>After UV disinfection: 0.7; 0.00 (0-1) (99% reduction)<br>The study demonstrated that UV disinfection was superior to manual cleaning for MRSA p<0.03). | SIGN **level 2+** evidence<br>McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Jinadatha 2014[4;16] |
| Meticillin resistant *Staphylococcus aureus* | Pulsed xenon UV | Hospital before and after study of impact of use of UV decontamination adjunct to traditional cleaning methods on discharge of selected rooms (intensive care unit for all discharges and | SIGN **level 3** evidence<br>McDonald-Arduino **Level V** – Demonstration of reduced microbial | Vianna et al 2016[14] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | transfers, but only *C.difficile* rooms for non-ICU discharges and transfers). Intervention resulted in a non-significant increase in MRSA rates in non-ICU areas (20%, p=0.23). There was a non-significant facility-wide increase in MRSA rates (50%, p=0.07). There was a 56% reduction in ICU rates, but this was not significant (p=0.22). Overall, there was a 29% facility-wide reduction in 3 infections (*C.difficile*, MRSA, VRE) in the intervention period compared to baseline (p=0.01). | pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | |
| Meticillin resistant *Staphylococcus aureus* | UV-C | Non-significant reduction in MRSA infection incidence rates from 0.39 in the baseline period (terminal cleaning only) to 0.38 in the intervention period (terminal cleaning + UV-C disinfection) (incidence rate change -1.2%, p=1). | SIGN **level 3** evidence<br>McDonald-Arduino **Level V** – Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | Napolitano et al 2015[12] |
| Oxacillin resistant *Staphylococcus aureus* | Disinfectant wipes (5 different wipes) | Study of efficacy of different wipes against bacteria on artificially contaminated keyboards. Wiping was performed for 5 seconds in a side to side manner on the key surfaces of the entire keyboard, 10 minutes of drying time was allowed.<br>Average CFU decrease after wiping (%):<br>Alcohol wipe: 95.29%<br>CaviWipes (QAC): 99.87%<br>Chlorine: 99.73%<br>Chlorox disinfecting (QAC): 100% | SIGN **level 3** evidence<br>McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Rutala et al 2006[75] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | Sani-Cloth Plus (QAC): 99.97%<br>Vesphene II SE: 99.90%<br>Sterile water: 98.58% | | |
| *Staphylococcus aureus* | Detergent wipes (7 different wipes) | The average removal of *S. aureus* from a stainless steel surface after a 10-second wipe was 1.45 $\log_{10}$ (range 0.24-3.25). Wipe D (Aquamed) removed significantly more *S.aureus* than the other wipes. All the wipes repeatedly transferred large number of *S.aureus* onto 3 consecutive surfaces except wipe G (Clinell new formulation), for which transfer of bacteria was below the detection limit of the test.<br>[Wipes performed better for removal and prevention of transfer for *A.baumanii* and *S.aureus* than *C.difficile* spores).] | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Ramm et al 2015[45] |
| Meticillin resistant *Staphylococcus aureus* | Disinfectant wipes (5 different wipes) | Study of decontamination of artificially contaminated plastic surfaces with wipes. Wipes swiped 1, 3 or 5 times over the surface (approximate contact time 1 second per swipe) and allowed to dry for 10 minutes.<br>For all 5 wipe types, swiping the surface 3 or 5 times eliminated more bacteria than only one swipe. Although not statistically significant, a reduction in the number of bacterial colonies was seen with 3 swipes of saline-moistened tissue compared with 1 swipe. At both 3 and 5 swipes for both MRSA, no type of wipe eliminated significantly more bacteria compared with the others. At one swipe, the chlorhexidine-alcohol wipes eliminated significantly more MRSA than the saline- | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Berendt et al 2011[19] |

| Pathogen | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | moistened wipe, and the quaternary ammonium compound wipe. | | |
| *Staphylococcus aureus* (meticillin susceptible and meticillin resistant) | Detergent and disinfectant wipes (2 different wipes) | After a 10-second wipe with an initial inoculum 6.09-6.93 $log_{10}$ CFU:<br>Detergent wipes removed 1.09- 2.49 $log_{10}$ CFU from dirty (with organic load) surfaces, and 1-2.26 $log_{10}$ CFU from clean surfaces.<br>Disinfectant (QAC) wipes removed 0.3-3.31 $log_{10}$ CFU from dirty surfaces and 0.97-3.31 $log_{10}$ CFU from clean surfaces.<br>Detergent wipes transferred bacteria (>100 CFU) to 8 consecutive agar plates. Disinfectant wipes also transferred bacteria to 8 consecutive agar plates (sometimes >100CFU).<br>Wipes were inoculated with 6.2–6.66 $log_{10}$ CFU with 10-seconds exposure to test bactericidal activity. Detergent wipes did not demonstrate any bactericidal effect. Disinfectant wipes demonstrated reductions of 1.57–3.42 and 1.66–5.55 $log_{10}$ CFU in the presence and absence of an organic load, respectively. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Williams et al 2009[57] |
| Meticillin resistant *Staphylococcus aureus* | Disinfectant Wipes (4 different wipes) | Study of decontamination of artificially contaminated tablet computers with wipes. All wipes were statistically better at removing bacteria from the tablet computer in comparison to the lint free cloth control. Clorox wipes (alkyldimethylbenzyl ammonium chloride 0.184%), Sani-Cloth CHG 2% (70% alcohol and 2% chlorhexidine) and Tristel Sporicidal wipes (0.1% - 0.12% chlorine dioxide) were the most effective. The Sani-cloth wipe was the only wipe found to exhibit a residual effect (no | SIGN **level 3** evidence McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Howell et al 2014[66] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | | growth after recontamination with microbes) for MRSA (and VRE). | | |
| | *Staphylococcus aureus* | Disinfectant Wipes (5 different wipes) | The three most effective wipes (2 0.5% accelerated H2O wipes; 1 sodium hypochlorite wipe with 1000ppm available chlorine) resulted in at least 7$log_{10}$ CFU reduction, least effective wipe resulted in >4$log_{10}$ reduction after a 10-second wipe. Three wipes resulted in subsequent transfer to another stainless steel surface. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Sattar et al 2015[51] |
| | *Staphylococcus aureus* | Disinfectant wipes (5 different wipes) | Although all 5 wipes removed all of the *S.aureus* from the surface of the anaesthesia machine, their performance was not statistically better than gauze with water. Only 1/5 wipes was outperformed by gauze and water when cleaning *S.aureus* from flat and ridged caps: 0.28%/17.2% Diisobutylphenoxyethoxyethyl dimethyl benzyl ammonium chloride wipe. The 0.5% hydrogen peroxide wipe was most effective at removing *S.aureus* from flat and ridged caps. [*S.aureus* was easier to remove than *C.sporogenes* and *B. atrophaeus* spores.] | SIGN **level 3** evidence McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Gonzalez et al 2015[65] |
| ***Staphylococcus epidermidis*** | *Staphylococcus epidermidis* | HINS | Inoculum: 100µL , $10^3$CFU/mL Complete inactivation after ~7 mins exposure to 405nm HINS with an average irradiance of 71mW/$cm^2$ Exposure to 5mW/cm2 for 1 hour reduced bacterial contamination by 83%. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | McDonald et al 2013[37] |
| | *Staphylococcus epidermidis* | HINS | Dose of 42 J/$cm^2$ resulted in 4.6 $log_{10}$ reduction | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory | MacLean et al 2009[35] |

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| | | | | demonstration of bioburden reduction efficacy | |
| | *Staphylococcus epidermidis* | Ozone | Inoculum: 20µL, 1 x $10^8$ CFU/ml. Exposure to 25ppm ozone for 30 min, with a 10 minute quench to remove residual ozone in a laboratory setting resulted in a 2.33 to 3.22 $log_{10}$ reduction (depending on material – highest log reduction on metal strip, lowest on cotton sheet). | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Moat et al 2009[39] |
| ***Streptococcus pneumoniae*** | *Streptococcus pneumoniae* | Disinfectant wipes (6 different wipes) | Surface of an anaesthesia machine inoculated with 7 x $10^5$ CFU *S pneumonia.* Wipe manufacturer's instructions for use were used, with 2 wipes per surface: one to remove gross contamination and one for disinfection (approx 3 seconds contact time).<br>Wipes:<br>**1** – ProSpray wipe<br>**2** – CleanCide Ready-to-use Germicidal detergent wipe<br>**3** – Clorox Germicidal Wipes<br>**4** – HypeWipe Bleach Towelette<br>**5** – Oxivir Tb Disinfectant Wipes<br>**6** – CaviWipe<br>All 6 wipes tested removed >98% initial inoculum. | SIGN **level 3** evidence McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Gold and Hitchins 2013[64] |
| ***Streptococcus pyogenes*** | *Streptococcus pyogenes* | HINS | Dose of 54 J/$cm^2$ resulted in 5 $log_{10}$ reduction | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | MacLean et al 2009[35] |

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| | *Streptococcus pyogenes* | Ozone | Inoculum: 20µL, 1 x $10^8$ CFU/ml. Exposure to 25ppm ozone for 30 min, with a 10 minute quench to remove residual ozone in a laboratory setting resulted in a 3.54 $log_{10}$ reduction on agar. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Moat et al 2009[39] |
| | *Streptococcus pyogenes* | Ozone | 25ppm ozone for 20 minutes followed by a short burst of humidity >90% resulted in ≥4.0 $log_{10}$ reduction | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Sharma and Hudson 2008[52] |
| ***Vibrio cholerae*** | *Vibrio cholerae* | AM silver | Minimum inhibitory concentration of colloidal silver nanoparticles against *V.cholerae* was found to be 6mg/L (contact time 24 hours). A concentration of 2-3mg/L was found to inhibit growth of *E.coli* in liquid suspension. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Le et al 2012[29] |
| ***Vibrio parahaemolyticus*** | *Vibrio parahaemolyticus* | Electrolysed water | Results similar to those for *Vibrio vulnificus* (see row below). | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Quan et al 2010[44] |
| ***Vibrio vulnificus*** | *Vibrio vulnificus* | Electrolysed water | Cell suspensions and cell cultures of *V.vulnificus* were treated for 30 s with sodium hypochlorite solution containing 35 mg/L available chlorine concentration (ACC) or weakly acidic electrolysed water (WAEW) containing 35 mg/L ACC. With an initial inoculum of 5.7 log CFU/mL, the number of viable *V.vulnificus* cells was reduced by 2.2 logs after treatment for 60s | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Quan et al 2010[44] |

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| | | | with sodium hypochlorite solution, while no cells survived treatment with WAEW for 30s. WAEW maintained its bactericidal activity for one week under open storage conditions, and for more than five weeks under closed storage conditions, demonstrating that it has a relatively stable shelf-life. | | |
| ***Yersinia pestis*** | *Yersinia pestis* | Chlorine dioxide gas | Complete activation of spores in 5/6 decontamination trials:<br>• 351ppm, 677ppm-hours, 50% RH;<br>• 377ppm, 890ppm-hours, 65% RH;<br>• 379ppm, 767ppm-hours, 65% RH;<br>• 385ppm, 770ppm-hours, 65% RH;<br>• 376ppm, 788ppm-hours, 64% RH.<br>[Initial inoculum $10^{10}$ CFU] | SIGN **level 3** evidence McDonald-Arduino **Level II –** Demonstration of in-use bioburden reduction effectiveness | Lowe et al 2013a[68] |
| **Gram-negative bacilli** | Gram-negative bacilli (including MDR strains) | HPV | Hospital-based (ICU) study comparing HPV decontamination with standard deep-cleaning (2000 ppm sodium hypochlorite for surfaces and 70% alcohol wipes for equipment) for the presence of environmental contamination with gram negative rods.<br>After deep-cleaning, but before HPV decontamination, 10/21 areas (47.6%) yielded gram negative rods. After HPV decontamination, no samples yielded Gram negative rods (p value not reported). | SIGN **level 3** evidence McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Otter et al 2010[71] |
| | Gentamicin resistant Gram-negative bacilli | HPV | 15 environmental samples taken from the hospital room of a patient with history of infection and colonisation with VRE, MRSA and gentamicin resistant Gram negative rods.<br>Proportion of sites positive for GNR:<br>• Before cleaning: 30% | SIGN **level 3** evidence McDonald-Arduino **Level III** – Demonstration of in-use bioburden reduction that may be clinically relevant | Otter et al 2007[73] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | | • After cleaning(detergent and QAC): 10%<br>• After HPV decontamination: 0<br>MRSA recontamination was identified 7 days after HPV decontamination. | | |
| | Extended spectrum β–lactamase (ESBL)-producing Gram-negative bacilli | HPV | At baseline, 0.82% samples from patient rooms contaminated with EBSL-GNR. After standard terminal cleaning, 0.96% of samples tested positive for EBSL-GNR. After HPV treatment, 0.13% of samples tested positive. This was a significant reduction compared to the proportion positive after standard terminal cleaning ($p<0.001$).<br>[HPV treatment: exposure to aerosolised 7% $H_2O_2$ solution with 0.25% peracetic acid and 30% acetic acid for a 30m minute contact time and 2 hour room ventilation period.] | SIGN **level 3** evidence<br>McDonald-Arduino **Level II** – Demonstration of in-use bioburden reduction effectiveness | Blazejewski et al 2015[61] |
| | Multidrug-resistant Gram-negative bacilli | HPV | The study demonstrated a reduced risk of acquisition of Gram-negative rods (individually) in patients admitted to rooms decontaminated using HPV compared with rooms decontaminated using standard methods, however this was not significant.<br>[The study looked at a number of multi-drug resistant organisms, and found that HPV decontamination was associated with a 64% lower risk of acquiring an MDRO overall (incidence rate ratio [IRR], 0.36; 95% confidence interval [CI], .19–.70; P < .001).] | SIGN **level 3** evidence<br>McDonald-Arduino **Level V –** Demonstration of reduced microbial pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | Passeretti et al 2013[13] |
| **Gram-negatives** | Multi-drug resistant gram negatives (MDR) | Pulsed xenon UV | Hospital before and after study of impact addition of UV disinfection to terminal decontamination on MDRO (and *C.difficile*) rates. MDRO rates decreased from 0.52/1000 | SIGN **level 3** evidence<br>McDonald-Arduino **Level V** – Demonstration of reduced microbial | Haas et al 2014[8] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
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| | | | patient days before the intervention to 0.42/1000 patient days during the intervention period (Rate ratio 0.81, (95% CI 0.66-0.98), p=0.04).<br>Limitation: simultaneous interventions occurring to reduce acquisition of MDROs and *C.difficile*, so cannot definitively attribute reductions to UV use. | pathogen acquisition (colonisation or infection) by patients via *non-outbreak* surveillance testing and clinical incidence | |
| ***Aspergillus niger*** | *Aspergillus niger* (spores (conidia); one melanised strain and one light-coloured mutant) | Ozone (± UV-C) | The melanised strain was more resistant to ozone than the strain without melanin. Conidia survival was lower when UV-C and ozone were used together compared to exposure to UV-C, demonstrating that ozone induced more inactivation in the presence of UV-C. Results showed that increasing the exposure time of ozone didn't reduce survival rates of the conidia. Ozone was demonstrated to be a less powerful agent for conidial inactivation than UV –C light, indicating that a combination of ozone and UV irradiation was necessary for optimal disinfection efficiency. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Liu et al 2014[31] |
| ***Aspergillus fumigatus*** | *Aspergillus fumigatus* | Steam – dry steam | Kill-time 7 minutes for inoculum with or without bovine serum albumen (initial inoculum $10^7$ CFU/mL). 7 minute kill time for 2.8% sodium hypochlorite with same inoculum concentration in the absence of BSA, but longer kill time required in presence of BSA (i.e. organic soil). | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Bagatinni et al 2015[17] |
| ***Cladosporium cladosporioides (allergen)*** | *Cladosporium cladosporioides* | Chlorine dioxide gas | Exposure to both 500ppm and 1000ppm chlorine dioxide gas for 24 hours completely inactivated *C.cladosporioides* (fungal colony inoculum 1 x $10^5$ conidia). | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of | Wilson et al 2005a[58] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
|---|---|---|---|---|---|
| | | | | bioburden reduction efficacy | |
| ***Candida parapsilosis*** | *Candida parapsilosis* | Steam – dry steam | Kill-time 7 minutes for inoculum with or without bovine serum albumen (initial inoculum $10^7$ CFU/mL). 7 minute kill time for 2.8% sodium hypochlorite with same inoculum concentration in the absence of BSA, but longer kill time required in presence of BSA (i.e. organic soil). | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Bagatinni et al 2015[17] |
| ***Chaetomium globosum*** **(allergen)** | *Chaetomium globosum* | Chlorine dioxide gas | For *C.globosum* colonies (inoculum 1 x $10^5$ conidia), exposure to 500ppm chlorine dioxide gas for 24 hours resulted in a treatment efficacy of 91%. At an exposure of 1,000 ppm for 24 hours, the treatment efficacy was 87%. *C.globosum* ascospores (inoculum 2 x $10^6$ conidia) were inactivated by 99.99% at both concentrations. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Wilson et al 2005a[58] |
| ***Penicillium chrysogenum*** | *Penicillium chrysogenum* | Chlorine dioxide gas | Exposure to both 500ppm and 1000ppm chlorine dioxide gas for 24 hours completely inactivated *P.chrysogenum* (fungal colony inoculum 1 x $10^5$ conidia). | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Wilson et al 2005a[58] |
| ***Stachybotrys chartarum*** | *Stachybotrys chartarum* | Chlorine dioxide gas | Exposure to both 500ppm and 1000ppm chlorine dioxide gas for 24 hours completely inactivated *S.chartarum* (fungal colony inoculum 1 x $10^5$ conidia).<br>The yeast toxicity assay results showed that chlorine dioxide did not detoxify the *S.chartarum* colonies. | SIGN **level 3** evidence McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Wilson et al 2005a[58] |
| **Hepatitis A virus** | Hepatitis A virus | Chlorine dioxide liquid | 4.30 (±0.18) $\log_{10}$ reduction after 5 minutes contact time at 22 ± 2°C for both 500ppm and | SIGN **level 3** evidence McDonald-Arduino **Level I** | Sabbah et al 2010[50] |

| Pathogen | | Technology | Results summary/[Notes] | SIGN Evidence level/McDonald hierarchy level | Study |
|---|---|---|---|---|---|
| | | | 1000ppm. | – Laboratory demonstration of bioburden reduction efficacy | |
| **Human norovirus (and surrogate: feline calicivirus)** | Norovirus and feline calicivirus | Ozone | Exposure to 20-25ppm ozone for 20 minutes was able to inactivate feline calicivirus by a factor of more than $10^3$, and in some cases beyond detection.<br>Virus dried onto hard surfaces (plastic, steel and glass), and soft surfaces such as fabric, cotton and carpet, were equally vulnerable to the treatment. | SIGN **level 3** evidence<br>McDonald-Arduino **Level I** – Laboratory demonstration of bioburden reduction efficacy | Hudson et al 2007[26] |

## Reference List


(1) SIGN 50 A guideline developer's handbook. 2011. Edinburgh, Scottish Intercollegiate Guidelines Network.
Ref Type: Generic

(2) McDonald LC, Arduino M. Editorial commentary: climbing the evidentiary hierarchy for environmental infection control. Clin Infect Dis 2013 Jan;56(1):36-9.

(3) Ghantoji SS, Stibich M, Stachowiak J, Cantu S, Adachi JA, Raad II, et al. Non-inferiority of pulsed xenon UV light versus bleach for reducing environmental *Clostridium difficile* contamination on high-touch surfaces in *Clostridium difficile* infection isolation rooms. Journal of Medical Microbiology 2015;64:191-4.

(4) Jinadatha C, Quezada R, Huber TW, Williams JB, Zeber JE, Copeland LA. Evaluation of a pulsed-xenon ultraviolet room disinfection device for impact on contamination levels of methicillin-resistant Staphylococcus aureus. BMC Infectious Diseases 2014;14:187.

(5) Carter Y, Barry D. Tackling C difficile with environmental cleaning. NURS TIMES 2011;107(36):2011-9.

(6) Gillespie E, Williams N, Sloane T, Wright L, Kotsanas D, Stuart RL. Using microfiber and steam technology to improve cleaning outcomes in an intensive care unit. AM J INFECT CONTROL 2015 Feb;43(2):177-9.

(7) Goldenberg SD, Patel A, Tucker D, French GL. Lack of enhanced effect of a chlorine dioxide-based cleaning regimen on environmental contamination with *Clostridium difficile* spores. J HOSP INFECT 2012;82:64-7.

(8) Haas JP, Menz J, Dusza S, Montecalvo MA. Implementation and impact of ultraviolet environmental disinfection in an acute care setting. AM J INFECT CONTROL 2014 Jun;42(6):586-90.

(9) Manian FA, Griesnauer S, Bryant A. Implementation of hospital-wide enhanced terminal cleaning of targeted patient rooms and its impact on endemic Clostridium difficile infection rates. AM J INFECT CONTROL 2013 Jun;41(6):537-41.

(10) Miller R, Simmons S, Dale C, Stachowiak J, Stibich M. Utilization and impact of a pulsed-xenon ultraviolet room disinfection system and multidisciplinary care team on Clostridium difficile in a long-term acute care facility. AM J INFECT CONTROL 2015;43(12):2015.

(11) Mitchell BG, Digney W, Locket P, Dancer SJ. Controlling methicillin-resistant Staphylococcus aureus (MRSA) in a hospital and the role of hydrogen peroxide decontamination: an interrupted time series analysis. BMJ open 2014;4(4):2014.

(12) Napolitano NA, Mahapatra T, Tang W. The effectiveness of UV-C radiation for facility-wide environmental disinfection to reduce health care–acquired infections. AM J INFECT CONTROL 2015 Dec;43(12):1342-6.

(13) Passaretti CL, Otter JA, Reich NG, Myers J, Shepard J, Ross T, et al. An evaluation of environmental decontamination with hydrogen peroxide vapor for reducing the risk of

patient acquisition of multidrug-resistant organisms. Clinical Infectious Diseases 2013;56(1):27-35.

(14) Vianna PG, Dale CR, Simmons S, Stibich M, Licitra CM. Impact of pulsed xenon ultraviolet light on hospital-acquired infection rates in a community hospital. AM J INFECT CONTROL 2016;44(3):01.

(15) Araujo EA, Andrade NJ, da Silva LH, Bernardes PC, de CTA, de Sa JP, et al. Antimicrobial effects of silver nanoparticles against bacterial cells adhered to stainless steel surfaces. J Food Prot 2012 Apr;75(4):701-5.

(16) Aydogan A, Gurol MD. Application of gaseous ozone for inactivation of Bacillus subtilis spores. Journal of the Air & Waste Management Association 2006 Feb;56(2):179-85.

(17) Bagattini M, Buonocore R, Giannouli M, Mattiacci D, Bellopede R, Grimaldi N, et al. Effect of treatment with an overheated dry-saturated steam vapour disinfection system on multidrug and extensively drug-resistant nosocomial pathogens and comparison with sodium hypochlorite activity. BMC Research Notes 2015;8(551).

(18) Baumann AR, Martin SE, Feng H. Removal of Listeria monocytogenes biofilms from stainless steel by use of ultrasound and ozone. Journal of Food Protection 2009 Jun;72(6):1306-9.

(19) Berendt AE, Turnbull L, Spady D, Rennie R, Forgie SE. Three swipes and you're out: How many swipes are needed to decontaminate plastic with disposable wipes? AM J INFECT CONTROL 2011;39(5):June.

(20) Chamakura K, Perez-Ballestro R, Luo Z, Bashir S, Liu J. Comparison of bactericidal activities of silver nanoparticles with common chemical disinfectants. Colloids & Surfaces B: Biointerfaces 2011;84(88):96.

(21) Chatuev BM, Peterson JW. Analysis of the sporicidal activity of chlorine dioxide disinfectant against Bacillus anthracis (Sterne strain). J HOSP INFECT 2010 Feb;74(2):178-83.

(22) Deza MA, Araujo M, Garrido MJ. Inactivation of Escherichia coli, Listeria monocytogenes, Pseudomonas aeruginosa and Staphylococcus aureus on stainless steel and glass surfaces by neutral electrolysed water. Letters in Applied Microbiology 2005;40(5):341-6.

(23) Friedline A, Zachariah M, Middaugh A, Heiser M, Khanna N, Vaishampayan P, et al. Sterilization of hydrogen peroxide resistant bacterial spores with stabilized chlorine dioxide. AMB Express 2015;5:24.

(24) Gant VA, Wren MWD, Rollins MSM, Jeanes A, Hickok SS, Hall TJ. Three novel highly charged copper-based biocides: Safety and efficacy against healthcare-associated organisms. Journal of Antimicrobial Chemotherapy 2007 Aug;60(2):294-9.

(25) Guffey JS, Payne W, James L. Inactivation of mycobacterium smegmatis following exposure to 405-nanometer light from a supraluminous diode array. Wounds-A Compendium of Clinical Research & Practice 2013 May;25(5):131-5.

(26) Hudson JB, Sharma M, Petric M. Inactivation of Norovirus by ozone gas in conditions relevant to healthcare. J HOSP INFECT 2007 May;66(1):40-5.

(27) Issa-Zacharia A, Kamitani Y, Tiisekwa A, Morita K, Iwasaki K. In vitro inactivation of Escherichia coli, Staphylococcus aureus and Salmonella spp. using slightly acidic electrolyzed water. Journal of Bioscience & Bioengineering 2010 Sep;110(3):308-13.

(28) Lawley TD, Clare S, Deakin LJ, Goulding D, Yen JL, Raisen C, et al. Use of purified Clostridium difficile spores to facilitate evaluation of health care disinfection regimens. Applied & Environmental Microbiology 2010 Oct;76(20):6895-900.

(29) Le AT, Le TT, Nguyen VQ, Tran HH, Dang DA, Tran QH, et al. Powerful colloidal silver nanoparticles for the prevention of gastrointestinal bacterial infections. Advances in Natural Sciences: Nanoscience and nanotechnology 2012;3.

(30) Li YJ, Zhu N, Jia HQ, Wu JH, Yi Y, Qi JC. Decontamination of Bacillus subtilis var. niger spores on selected surfaces by chlorine dioxide gas. Journal of Zhejiang University SCIENCE B 2012 Apr;13(4):254-60.

(31) Liu J, Zhou L, Chen JH, Mao W, Li WJ, Hu W, et al. Role of ozone in UV-C disinfection, demonstrated by comparison between wild-type and mutant conidia of Aspergillus niger. Photochemistry & Photobiology 2014 May;90(3):615-21.

(32) Lowe J, Gibbs S, Iwen P, Smith P. A Case Study on Decontamination of a Biosafety Level-3 Laboratory and Associated Ductwork Within an Operational Building Using Gaseous Chlorine Dioxide. Journal of Occupational & Environmental Hygiene 2012 Dec;9(12):D196-D205.

(33) Luna VA, Hall TJ, King DJ, Cannons AC. Susceptibility of 169 USA300 methicillin-resistant Staphylococcus aureus isolates to two copper-based biocides, CuAL42 and CuWB50. Journal of Antimicrobial Chemotherapy 2010;65:939-41.

(34) Maclean M, Macgregor SJ, Anderson JG, Woolsey G. High-intensity narrow-spectrum light inactivation and wavelength sensitivity of Staphylococcus aureus. FEMS Microbiology Letters 2008 Aug;285(2):227-32.

(35) Maclean M, Macgregor SJ, Anderson JG, Woolsey G. Inactivation of bacterial pathogens following exposure to light from a 405-nanometer light-emitting diode array. Applied & Environmental Microbiology 2009 Apr;75(7):1932-7.

(36) Maclean M, Murdoch LE, Macgregor SJ, Anderson JG. Sporicidal effects of high-intensity 405 nm visible light on endospore-forming bacteria. Photochemistry & Photobiology 2013 Jan;89(1):120-6.

(37) McDonald RS, Gupta S, Maclean M, Ramakrishnan P, Anderson JG, Macgregor SJ, et al. 405 nm Light exposure of osteoblasts and inactivation of bacterial isolates from arthroplasty patients: potential for new disinfection applications? European Cells & Materials 2013;25:204-14.

(38) McKenzie K, Maclean M, Timoshkin IV, Endarko E, Macgregor SJ, Anderson JG. Photoinactivation of bacteria attached to glass and acrylic surfaces by 405 nm light: potential application for biofilm decontamination. Photochemistry & Photobiology 2013 Jul;89(4):927-35.

(39) Moat J, Cargill J, Shone J, Upton M. Application of a novel decontamination process using gaseous ozone. Canadian Journal of Microbiology 2009 Aug;55(8):928-33.

(40) Murdoch LE, Maclean M, Macgregor SJ, Anderson JG. Inactivation of Campylobacter jejuni by exposure to high-intensity 405-nm visible light. Foodborne Pathogens & Disease 2010 Oct;7(10):1211-6.

(41) Murdoch LE, Maclean M, Endarko E, Macgregor SJ, Anderson JG. Bactericidal effects of 405nm light exposure demonstrated by inactivation of Escherichia, Salmonella, Shigella, Listeria, and Mycobacterium species in liquid suspensions and on exposed surfaces. Thescientificworldjournal 2012;2012:137805.

(42) Nicholas R, Dunton P, Tatham A, Fielding L. The effect of ozone and open air factor on surface-attached and biofilm environmental Listeria monocytogenes. Journal of Applied Microbiology 2013 Aug;115(2):555-64.

(43) Perez J, Springthorpe VS, Sattar SA. Activity of selected oxidizing microbicides against the spores of Clostridium difficile: relevance to environmental control. AM J INFECT CONTROL 2005 Aug;33(6):320-5.

(44) Quan Y, Choi KD, Chung D, Shin IS. Evaluation of bactericidal activity of weakly acidic electrolyzed water (WAEW) against Vibrio vulnificus and Vibrio parahaemolyticus. International Journal of Food Microbiology 2010 Jan 1;136(3):255-60.

(45) Ramm L, Siani H, Wesgate R, Maillard J-Y. Pathogen transfer and high variability in pathogen removal by detergent wipes. AM J INFECT CONTROL 2015;43(7):01.

(46) Rastogi VK, Wallace L, Smith LS, Ryan SP, Martin B. Quantitative method to determine sporicidal decontamination of building surfaces by gaseous fumigants, and issues related to laboratory-scale studies. Applied & Environmental Microbiology 2009 Jun;75(11):3688-94.

(47) Rastogi VK, Ryan SP, Wallace L, Smith LS, Shah SS, Martin GB. Systematic evaluation of the efficacy of chlorine dioxide in decontamination of building interior surfaces contaminated with anthrax spores. Applied & Environmental Microbiology 2010 May;76(10):3343-51.

(48) Rutala WA, Gergen MF, Weber DJ. Efficacy of different cleaning and disinfection methods against clostridium difficile spores: Importance of physical removal versus sporicidal inactivation. Infection Control and Hospital Epidemiology 2012;33(12):December.

(49) Ryan SP, Lee SD, Calfee MW, Wood JP, McDonald S, Clayton M, et al. Effect of inoculation method on the determination of decontamination efficacy against Bacillus spores. World Journal of Microbiology & Biotechnology 2014 Oct;30(10):2609-23.

(50) Sabbah S, Springthorpe S, Sattar SA. Use of a mixture of surrogates for infectious bioagents in a standard approach to assessing disinfection of environmental surfaces. Applied & Environmental Microbiology 2010 Sep;76(17):6020-2.

(51) Sattar SA, Bradley C, Kibbee R, Wesgate R, Wilkinson MA, Sharpe T, et al. Disinfectant wipes are appropriate to control microbial bioburden from surfaces: use of a new ASTM standard test protocol to demonstrate efficacy. J HOSP INFECT 2015 Dec;91(4):319-25.

(52) Sharma M, Hudson JB. Ozone gas is an effective and practical antibacterial agent. AM J INFECT CONTROL 2008 Oct;36(8):559-63.

(53) Siani H, Cooper C, Maillard J-Y. Efficacy of "sporicidal" wipes against Clostridium difficile. AM J INFECT CONTROL 2011;39(3):April.

(54) Song L, Wu J, Xi C. Biofilms on environmental surfaces: evaluation of the disinfection efficacy of a novel steam vapor system. AM J INFECT CONTROL 2012 Dec;40(10):926-30.

(55) Speight S, Moy A, Macken S, Chitnis R, Hoffman PN, Davies A, et al. Evaluation of the sporicidal activity of different chemical disinfectants used in hospitals against Clostridium difficile. J HOSP INFECT 2011 Sep;79(1):18-22.

(56) Trajtman AN, Manickam K, Alfa MJ. Microfiber cloths reduce the transfer of Clostridium difficile spores to environmental surfaces compared with cotton cloths. AM J INFECT CONTROL 2015;43(7):01.

(57) Williams GJ, Denyer SP, Hosein IK, Hill DW, Maillard J-Y. Limitations of the efficacy of surface disinfection in the healthcare setting. Infection Control and Hospital Epidemiology 2009;30(6):June.

(58) Wilson SC, Wu C, Andriychuk LA, Martin JM, Brasel TL, Jumper CA, et al. Effect of chlorine dioxide gas on fungi and mycotoxins associated with sick building syndrome. Applied & Environmental Microbiology 2005 Sep;71(9):5399-403.

(59) Zoutman D, Shannon M, Mandel A. Effectiveness of a novel ozone-based system for the rapid high-level disinfection of health care spaces and surfaces. AM J INFECT CONTROL 2011 Dec;39(10):873-9.

(60) Barbut F, Menuet D, Verachten M, Girou E. Comparison of the efficacy of a hydrogen peroxide dry-mist disinfection system and sodium hypochlorite solution for eradication of clostridium difficile spores. Infection Control and Hospital Epidemiology 2009 Jun;30(6):507-14.

(61) Blazejewski C, Wallet F, Rouze A, Le GR, Ponthieux S, Salleron J, et al. Efficiency of hydrogen peroxide in improving disinfection of ICU rooms. Critical Care (London, England) 2015;19:30.

(62) Doan L, Forrest H, Fakis A, Craig J, Claxton L, Khare M. Clinical and cost effectiveness of eight disinfection methods for terminal disinfection of hospital isolation rooms contaminated with Clostridium difficile 027. J HOSP INFECT 2012 Oct;82(2):114-21.

(63) French GL, Otter JA, Shannon KP, Adams NMT, Watling D, Parks MJ. Tackling contamination of the hospital environment by methicillin-resistant Staphylococcus aureus (MRSA): A comparison between conventional terminal cleaning and hydrogen peroxide vapour decontamination. J HOSP INFECT 2004 May;57(1):31-7.

(64) Gold KM, Hitchins VM. Cleaning assessment of disinfectant cleaning wipes on an external surface of a medical device contaminated with artificial blood or Streptococcus pneumoniae. AM J INFECT CONTROL 2013;41(10):October.

(65) Gonzalez EA, Nandy P, Lucas AD, Hitchins VM. Ability of cleaning-disinfecting wipes to remove bacteria from medical device surfaces. AM J INFECT CONTROL 2015;43(12):1331-5.

(66) Howell V, Thoppil A, Mariyaselvam M, Jones R, Young H, Sharma S, et al. Disinfecting the iPad: evaluating effective methods. J HOSP INFECT 2014;87:77-83.

(67) Lowe J, Hewlett A, Iwen P, Smith P, Gibbs S. Evaluation of Ambulance Decontamination Using Gaseous Chlorine Dioxide. Prehospital Emergency Care 2013 Jul;17(3):401-8.

(68) Lowe J, Gibbs S, Iwen P, Smith P, Hewlett A. Decontamination of a Hospital Room Using Gaseous Chlorine Dioxide: Bacillus anthracis, Francisella tularensis, and Yersinia pestis. Journal of Occupational & Environmental Hygiene 2013 Oct;10(10):533-9.

(69) Maclean M, Macgregor SJ, Anderson JG, Woolsey GA, Coia JE, Hamilton K, et al. Environmental decontamination of a hospital isolation room using high-intensity narrow-spectrum light. J HOSP INFECT 2010 Nov;76(3):247-51.

(70) Maclean M, Booth M, Anderson J, MacGregor S, Woolsey G, Coia J, et al. Continuous decontamination of an intensive care isolation room during patient occupancy using 405 nm light technology. Journal of Infection Prevention 2013 Sep;14(5):176-81.

(71) Otter JA, Yezli S, Schouten MA, van Zanten AR, Houmes-Zielman G, Nohlmans-Paulssen MK. Hydrogen peroxide vapor decontamination of an intensive care unit to remove environmental reservoirs of multidrug-resistant gram-negative rods during an outbreak. AM J INFECT CONTROL 2010 Nov;38(9):754-6.

(72) Stibich M, Stachowiak J, Tanner B, Berkheiser M, Moore L, Raad I, et al. Evaluation of a pulsed-xenon ultraviolet room disinfection device for impact on hospital operations and microbial reduction. INFECT CONTROL HOSP EPIDEMIOL 2011 Mar;32(3):286-8.

(73) Otter JA, Cummins M, Ahmad F, van TC, Drabu YJ. Assessing the biological efficacy and rate of recontamination following hydrogen peroxide vapour decontamination. J HOSP INFECT 2007 Oct;67(2):182-8.

(74) Orenstein R, Aronhalt KC, McManus Jr JE, Fedraw LA. A targeted strategy to wipe out *Clostridium difficile*. Infection Control and Hospital Epidemiology 2011;32(11):1137-9.

(75) Rutala WA, White MS, Gergen MF, Weber DJ. Bacterial contamination of keyboards: efficacy and functional impact of disinfectants. INFECT CONTROL HOSP EPIDEMIOL 2006 Apr;27(4):372-7.

(76) Lowe JJ, Gibbs SG, Iwen PC, Smith PW, Hewlett AL. Impact of chlorine dioxide gas sterilization on nosocomial organism viability in a hospital room. International Journal of Environmental Research & Public Health [Electronic Resource] 2013 Jun;10(6):2596-605.