Supplementary Material

Supplementary Methods

Method S1: Effect of Drying Time on the Recovery of Microorganisms from Cotton

Sterile 25cm^2 swatches of 100% cotton textile were inoculated with 500 µL test suspension (8 or 2 $\log_{10}$ CFU/ml). After 0, 3, 6 and 18 h drying at room temperature (~21°C), samples were placed in 30 ml PBS-T and shaken by hand 30 times. The supernatant was spiral plated, spread plated or membrane filtered onto nutrient agar and resulting colonies were enumerated after 24 h incubation at 37°C.

Method S2: Disinfectant Suspension Test

The $\log_{10}$ reduction of *E. faecium* was investigated using the BS EN 1040:2005 disinfectant suspension test (British Standards Institution, 2005). Disinfectant solutions (8 ml) were mixed with 1 ml water and 1 ml *E. faecium* (1.5-5x10^8 CFU/ml) and incubated at room temperature for 13 minutes. After incubation, 1 ml test solution was added to 8 ml neutraliser with 1 ml water and incubated at room temperature for 5 minutes. The neutraliser solution was spread or spiral plated in duplicate onto nutrient agar and *E. faecium* was enumerated after both 24- and 48-hours' incubation at 37°C. A control of *E. faecium* in water only was included.

Method S3: Swatch Suspension Test

An adapted BS EN 1040:2005 method using swatches inoculated with *E. faecium* was thus developed to identify sublethal concentrations of the disinfectants. Sterile cotton swatches (1 cm^2) were inoculated with 20 µl *E. faecium* (10^8 CFU/ml) and allowed to dry overnight at room temperature. Swatches were immersed in 10 ml disinfectant solution and incubated at room temperature for 13 minutes without agitation. The swatch was then transferred to 9 ml neutraliser solution, shaken by hand 30 times and incubated at room temperature for 5 minutes. Neutraliser solutions were then plated onto nutrient agar and enumerated after both 24 and 48 hours' incubation at 37°C. A water only control was included throughout.

Method S4: Swatch-Based Neutraliser Efficacy Validation Test

A neutraliser validation method was adapted from the BS EN 1040:2005 neutralisation-dilution validation tests (British Standards Institution, 2005), using inoculated cotton swatches in place of microbial suspensions. Cotton swatches (1 cm^2) were inoculated with *Enterococcus faecium* (10^6 CFU/swatch) and allowed to dry overnight at room temperature. The starting inoculum was determined by shaking untreated swatches 30 times in 30 ml phosphate buffered saline with 2 g/l tween 80 (untreated control). Samples were spread plated on nutrient agar and enumerated after 48 hours' incubation at 37°C.

Neutraliser Toxicity

The neutraliser toxicity was determined by shaking swatches in 30 ml neutraliser 30 times prior to 5 minutes incubation at room temperature. Surviving microorganisms were spread plated on nutrient agar and enumerated after 48 hours' incubation at 37°C. An experimental conditions control (30 ml water only) was included.

The neutraliser was considered non-toxic where the $\log_{10}$ CFU/ml reduction compared to untreated controls was less than or equal to 0.5, according to BS ISO 18184:2019 (British Standards Institution, 2019; equation 1):

Log_{10} CFU/ml (untreated control) - $\log_{10}$ CFU/ml (test) ≤ 0.5 (Equation 1)

Neutraliser Efficacy

Aliquots (1 ml) of peracetic acid (1.2 ml/L) , sodium hypochlorite (2 ml/L) and sodium dodecyl sulfate (SDS; 10% w/v) were mixed with 29 ml neutraliser by vortexing and incubated for 5 minutes at room temperature. A water only control was included. *E. faecium*-inoculated swatches were then added to the neutralising solutions and shaken by hand 30 times. The samples were incubated for 30 minutes at room temperature, and surviving microorganisms were enumerated on nutrient agar as described above.

The neutraliser was considered effective where the $\log_{10}$ CFU/ml reduction compared to the water only control was less than or equal to 0.5, according to BS ISO 18184:2019 (British Standards Institution, 2019; equation 2):

Log_{10} CFU/ml (water only control) - $\log_{10}$ CFU/ml (test) ≤ 0.5 (Equation 2)

Results were compared to that of the BS EN 1040:2005 neutraliser toxicity and efficacy tests performed using the BS EN 1040:2005 method with suspensions of *E. faecium*, which determined that the neutraliser was non-toxic and effective against peracetic acid, sodium hypochlorite and SDS.

Method S5: Assessment of the Stability of the Polyethersulphone Membrane through a Wash System

PES membranes were laundered in an extractor washer with non-ionic surfactants at 75°C, for 30 min and then tumbled dried at 90°C for 60 min. Tensile strength and visual integrity of the membrane were observed pre and post laundering. Flow rate pre and post laundering was also observed.

Supplementary Data

Effect of Drying Time on Recovery of Microorganisms from Cotton

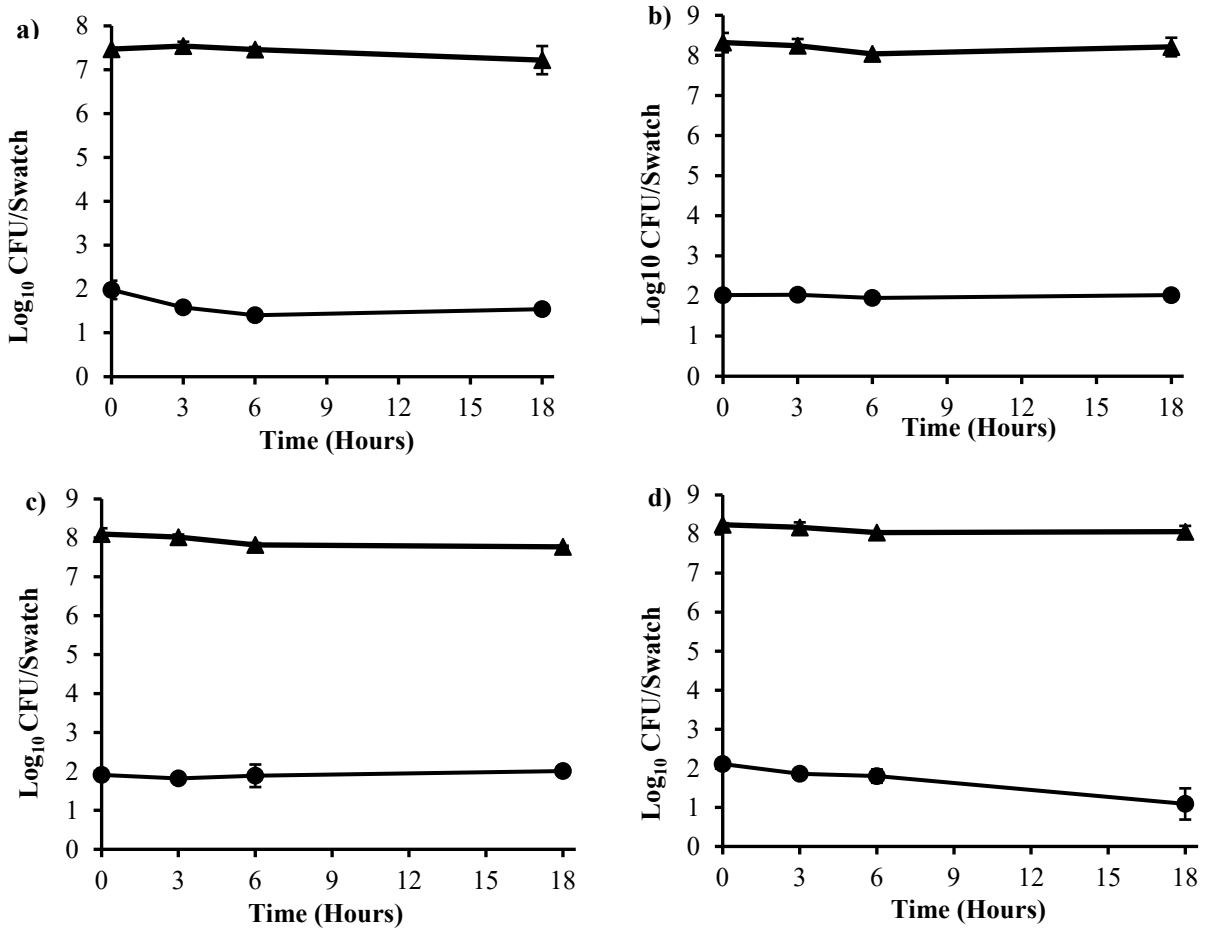

Figure S1: Log_{10} CFU/swatch recovery of (a) *B. cereus* spores, (b) *E. faecium,* (c) *E. coli* and (d) *S. aureus* following incubation at room temperature (n = 4 ± standard error of the mean [SEM]). ▲ 10^8 CFU/swatch inoculum; ● 10^2 CFU/swatch inoculum.

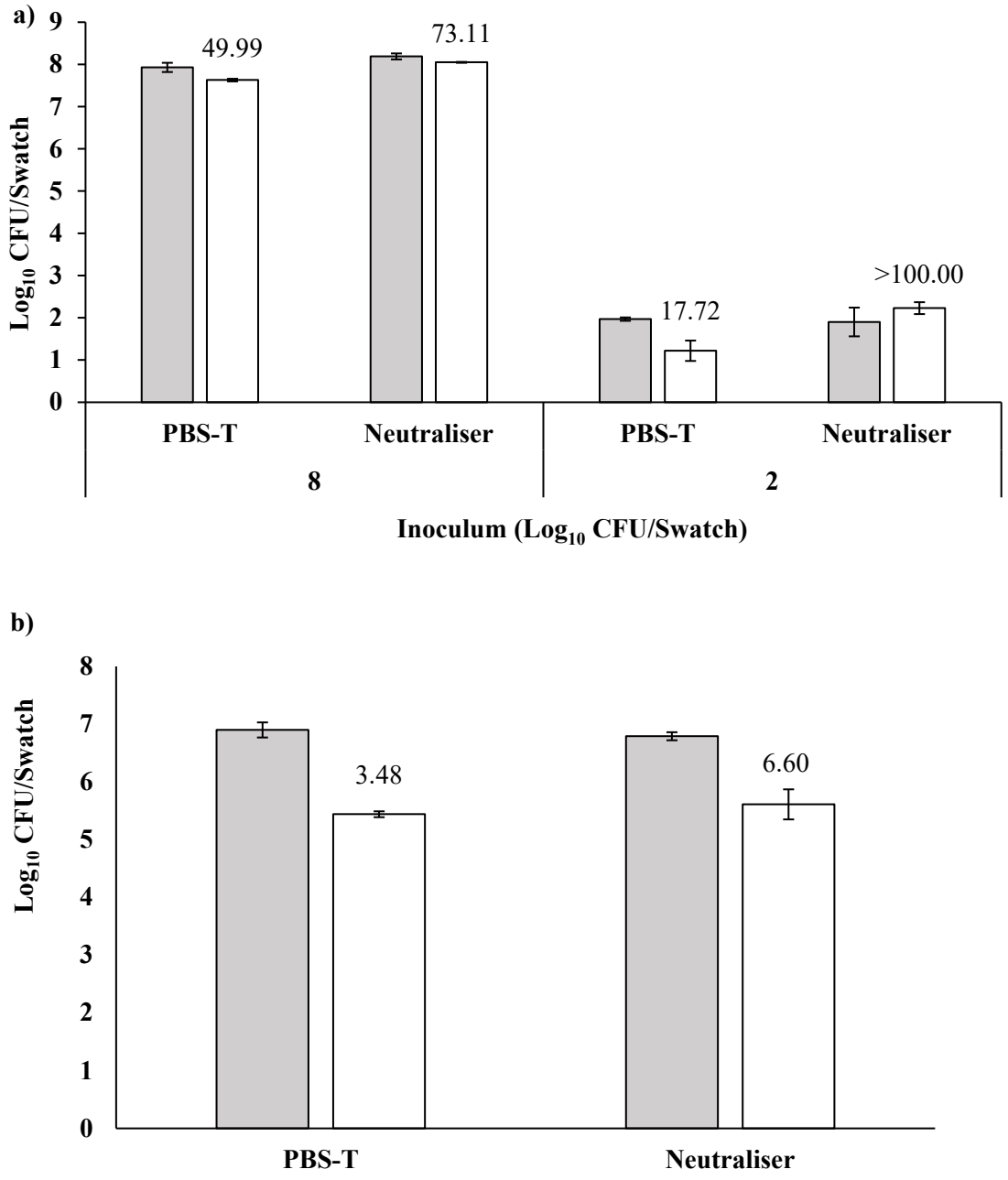

Figure S2: Log_{10} CFU/swatch recovery of *E. faecium* (8 and 2 log_{10} CFU/swatch) from (a) 25 cm^2 and (b) 1 cm^2 cotton swatches (6 log_{10} CFU/swatch) when shaken by hand in 30 ml neutraliser in comparison to PBS-T. Closed bars, Original inoculum; open bars, recovery from swatch. Percentage recoveries from the original inocula are shown as data labels.

Chemical Susceptibility of *E. faecium*

Table S1: Log_{10} reduction of *E. faecium* (BS EN 1040:2005, 8.02 log_{10} CFU; swatch test, 6.78 log_{10} CFU) by a range of disinfectants and detergents according to both the BS EN 1040:2005 suspension test and adapted swatch suspension test, and times difference in the concentration of disinfectant required to produce an approximately equivalent log_{10} reduction.

Disinfectant	BS EN 1040:2005			Swatch Test				Concentration Difference
	Concentration (ml/L)	Log_{10} CFU	Log_{10} Reduction from Inoculum	Concentration (ml/L)	Log_{10} CFU	Log_{10} Reduction from Inoculum	Log_{10} Reduction from Water	
Water only	-	2.85 ± 0.09*	0.08	-	6.53 ± 0.20	0.35	-	-
Peracetic Acid	0.028	3.49 ± 0.68	3.84	0.64	3.58	3.33 ± 0.02	3.58	22.86
Sodium Hypochlorite	0.030	4.00 ± 0.78	3.54	0.80	3.25	3.66 ± 0.22	3.25	26.67
BAC	0.023	4.05 ± 0.37	3.65	0.093	3.82 ± 0.49	3.09	3.24	4.11
DDAC	0.028	4.22 ± 0.51	3.48	0.150	3.48 ± 0.55	3.10	3.24	5.45
Hydrogen Peroxide	5.860	3.45 ± 0.15	3.91	4.690	3.58 ± 0.07	3.38	2.95	0.80
Hypochlorous Acid	0.340	4.69 ± 0.24	3.63	3.130	3.68 ± 0.06	3.07	2.84	9.19
SDS	100.00	4.92 ± 0.22	2.92	0.600	2.79 ± 0.36 §	3.97	3.74	166.67

***** BS EN 1040:2005 water control conducted using an inoculum of 2.93 log_{10} CFU/ml as per test validation requirements. § Membrane filtration of swatch test supernatant; 0.41 ± 0.41 log_{10} CFU recovery (3 of 4 repeats negative for growth.

Table S2: Log_{10} CFU/swatch recovery of *E. faecium* from cotton swatches after treatment in solution without a membrane compared to an industrial wash without temperature. * Significant ($p \leq 0.05$) difference between loose swatch in solution and in the wash. § Significant ($p \leq 0.05$) reduction compared to the water only control.

Disinfectant	**Treatment**	**Log_{10} Recovery**		**Log Reduction from Inoculum**	**Log Reduction from Water Only Control**
		Pre-Treatment	**Post-Treatment**		
Water Only Control	Loose swatch (solution)	6.21 ± 0.13	6.36 ± 0.14	-0.14	-
	Loose swatch (laundered)	5.46 ± 0.13	3.63 ± 0.08	1.83*	-
Reference Detergent	Loose swatch (solution)	6.76 ± 0.06	6.43 ± 0.10	0.33	0.47
	Loose swatch (laundered)	6.13 ± 0.31	1.27 ± 0.35	4.86*	3.03§

Swatch-Based Neutraliser Efficacy Validation Test

Table S3: Comparison of swatch-based neutraliser validation tests to BS EN 1040:2005 suspension test against *E. faecium* (n = 4 ± SEM).

Condition	Disinfectant	Swatch Test			BS EN 1040:2005	
		Log_{10} CFU/ml	Log_{10} Reduction	Pass/Fail*	CFU/ml	Pass/Fail†
Untreated Control	-	4.92 ± 0.21	-	-	82.60 ± 6.81	-
Experimental Conditions Control	-	4.90 ± 0.23	0.02	Pass	72.63 ± 10.10	Pass
Neutraliser Toxicity	-	4.60 ± 0.26	0.32	Pass	69.38 ± 10.39	Pass
Neutraliser Efficacy	Water Only	4.74 ± 0.18	0.18	-	-	-
	Peracetic Acid	4.78 ± 0.15	-0.04	Pass	69.75 ± 10.68	Pass
	Sodium Hypochlorite	4.71 ± 0.20	0.03	Pass	74.13 ± 10.38	Pass
	SDS	4.33 ± 0.41	0.41	Pass	56.38 ± 6.03	Pass

*Pass, <0.5 log_{10} CFU/ml reduction from untreated control (experimental conditions control and neutraliser toxicity) or water only control (neutraliser efficacy);

†Pass, >0.5 CFU/ml remaining compared to the untreated control.

Chemical susceptibility

Table S4: Log_{10} CFU/swatch survival of *Enterococcus* sp. treated with disinfectants for 13 min according to the adapted swatch suspension test (n = 4 ± SEM). Log_{10} reduction from water is shown in brackets.

		Log_{10} CFU/swatch (Log_{10} reduction)	
Disinfectant	**Concentration**	***E. faecium***	***E. hirae***
Water	-	6.53 ± 0.20	5.83 ± 0.43
Peracetic acid	1.2 ml/L	0.00 ± 0.00 (6.53)	0.00 ± 0.00 (6.53)
	0.64 ml/L	3.33 ± 0.02 (3.20)	0.00 ± 0.00 (6.53)
Chlorine	2 ml/L	0.00 ± 0.00 (6.53)	0.35 ± 0.32 (6.18)
	0.8 ml/L	3.66 ± 0.22 (2.87)	0.58 ± 0.45 (5.95)
SDS	1 % w/v	0.00 ± 0.00 (6.53)	0.00 ± 0.00 (6.53)
	0.06 % w/v	2.79 ± 0.38 (3.74)	≥3.00 ± 0.49 (≤3.53)