

Evaluation of the surface disinfection potential of the AIRsteril device



Dr Louise Fletcher

Areas of expertise: waste management; aerobiology; environmental microbiology

CONTENTS

| 1. Surface disinfection potential2 |
|---|
| 1.1 Objectives of the Study2 |
| 1.2 Test microorganisms2 |
| 1.3 Culture preparation2 |
| 1.4 Preparation of stainless steel squares2 |
| 1.5 Surface experiment methodology |
| 1.6 Enumeration of the bacteria on the steel squares4 |
| 1.7 Results |





1.1 Objectives of the Study

The objective of the experiments was to determine the efficacy of the device in terms of its ability to reduce the concentration of viable microorganisms on stainless steel surfaces.

1.2 Test microorganisms

The surface tests were carried out using pure cultures of two microorganisms as follows: Staphylococcus aureus – ATCC6538 Escherichia coli - ATCC10536 Clostridium difficile

1.3 Culture preparation

The S. aureus and E. coli cultures were prepared by using the cultiloops to inoculate 50ml of sterile nutrient broth (Oxoid, UK). The broths were then incubated at 37°C for 24 hours and shaken at 100rpm. After incubation the culture was enumerated and then used to inoculate the surfaces. The C. diffcile culture was prepared by inoculating a 50ml sterile nutrient broth (previously purged with nitrogen gas to remove any oxygen before autoclaving) with a small aliquot of stock laboratory culture and incubating for 24 hours and shaken at 100rpm. Because C. difficile is an anaerobic bacteria the culture was prepared in sealed Wheaton bottles and inoculated using a sterile syringe through the rubber septum

1.4 Preparation of stainless steel squares

Prior to use the stainless steel squares were placed in a beaker and washed in detergent for 1 hour after which they were rinsed with deionised water. Each square was then washed individually for 10 seconds with deionised water to make sure there was no residual detergent. The squares were then autoclaved at 121°C for 15 minutes.

1.5 Surface experiment methodology

The tests were undertaken using previously decontaminated stainless steel squares prepared as outlined above. The squares were inoculated respectively with 50 l of a pure culture of S. aureus, E. coli C. difficile. The squares were placed into a laminar flow microbiological cabinet until the inoculum had completely dried. While the squares were being prepared the ventilation system in the chamber was switched on and operated at 12 AC/hr to purge the air inside the chamber after which it was switched off. The device was placed into the chamber along with a small support for the squares which was within 1m of the device and the device was switched on and operated for 2 hours before the start of the test. After drying, 15 of the squares inoculated for each microorganism were placed into the aerobiological chamber and 5 were retained for immediate enumeration. The test squares were then exposed to the test device and at 8 hours, 24 hours and 48 hours 5 squares for each microorganism were removed from the chamber.



INOCULATED STEEL SQUARES



POSITIONING OF THE DEVICE AND STEEL SQUARES IN THE AEROBIOLOGY CHAMBER

1.6 Enumeration of the bacteria on the steel squares

After exposure the squares were removed from the chamber and prepared for analysis. The surface of each square was swabbed using a sterile swab soaked in sterile ringer's solution. The end of the swab was then snapped off and placed into a small bottle containing 10ml of sterile ringer's solution. Each bottle was then shaken for 30 minutes and vortexed for 1 minute. The solution was then diluted as required and plated out onto sterile tryptone soya agar plates. All the plates were then incubated for 24 hours at 37°C after which the total number of colonies on each plate were counted. The colony counts were then used to calculate the concentration of microorganisms in the 10ml of ringer's solution and therefore the number of microorganisms recovered from each square surface. The counts from the five replicate steel squares were then used to determine the mean concentration with and without exposure to the device and this data was used to determine the mean reduction as a percentage.

1.7 Results

Table 1 shows the results obtained during the surface exposure experiment carried out using E. coli. The initial mean concentration on the steel squares was 10280 cfu which was reduced to 2100 cfu after exposure for 8 hours, 240 cfu after 24 hours and 10 cfu after 48 hours. This represents a reduction in the number of E. coli on the steel squares after 8 hours, 24 hours and 48 hours of 79.6%, 97.7% and 99.9% respectively.

| Square | Initial | 8 hours | 24 hours | 48 hours |
|-----------|---------|---------|----------|----------|
| 1 | 5900 | 2650 | 100 | 0 |
| 2 | 9200 | 1700 | 250 | 50 |
| 3 | 14600 | 1250 | 350 | 0 |
| 4 | 13000 | 3000 | 50 | 0 |
| 5 | 8700 | 1900 | 450 | 0 |
| Mean | 10280 | 2100 | 240 | 10 |
| SD | 3497 | 713 | 167 | 22 |
| Reduction | - | 79.6% | 97.7% | 99.9% |

TABLE 1 RESULTS OF THE SURFACE EXPOSURE EXPERIMENT CARRIED OUT USING E. COLI

As can be seen in Table 2 a similar trend was observed with S. aureus with an initial mean concentration of S. aureus on the steel squares slightly higher than that of E. coli at 14360 cfu. This was reduced to 1810 cfu after exposure for 8 hours, 217 cfu after 24 hours and 84 cfu after 48 hours. This represents a reduction in the number of S. aureus on the steel squares after 8 hours, 24 hours and 48 hours of 87.4%, 91.1% and 99.5% respectively.

| Square | Initial | 8 hours | 24 hours | 48 hours |
|-----------|---------|---------|----------|----------|
| 1 | 12300 | 1400 | 1450 | 200 |
| 2 | 14100 | 1050 | 1050 | 0 |
| 3 | 15200 | 2450 | 1500 | 0 |
| 4 | 18500 | 1750 | 1050 | 50 |
| 5 | 11700 | 2400 | 1350 | 100 |
| Mean | 14360 | 1810 | 1280 | 70 |
| SD | 2703 | 614 | 217 | 84 |
| Reduction | - | 87.4% | 91.1% | 99.5% |

TABLE 2 RESULTS OF THE SURFACE EXPOSURE EXPERIMENT CARRIED OUT USING S. AUREUS

Table 3 shows the results of the surface exposure experiment carried out using C. difficile. The initial mean concentration of C. difficile on the steel squares slightly lower than that of either the E. coli or the S. aureus at 9460 cfu. This was reduced to 810 cfu after exposure for 8 hours, 180 cfu after 24 hours and 40 cfu after 48 hours. This represent a reduction in the number of C. difficile on the steel squares after 8 hours, 24 hours and 48 hours of 91.44%, 98.1% and 99.6% respectively.

| Square | Initial | 8 hours | 24 hours | 48 hours |
|-----------|---------|---------|----------|----------|
| 1 | 10300 | 1300 | 250 | 50 |
| 2 | 5100 | 1450 | 100 | 0 |
| 3 | 10700 | 650 | 350 | 100 |
| 4 | 8100 | 300 | 150 | 50 |
| 5 | 13100 | 350 | 50 | 0 |
| Mean | 9460 | 810 | 180 | 40 |
| SD | 3015 | 535 | 120 | 42 |
| Reduction | - | 91.4% | 98.1% | 99.6% |

TABLE 3 RESULTS OF THE SURFACE EXPOSURE EXPERIMENT CARRIED OUT USING C. DIFFICILE