

**Biocidal Surface Test  
- Clinell Wipes**

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**Project Report Prepared for GAMA Healthcare Ltd**

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## Summary

The ability of Clinell wipes to disinfect contaminated steel surfaces was assessed using hospital grade stainless steel discs (9cm dia) pre-contaminated with an *E. coli* broth. These discs were air dried and then wiped once with a Clinell wipe, contact time between the wipe and the surface was between 2 and 5 seconds. The discs were left for 5 minutes before the surfaces were swabbed with pre-moistened swabs to recover any remaining viable bacteria. The effectiveness of the wipes was assessed by comparison with un-wiped control discs contaminated in the same manner as the test discs.

After air drying the discs retained on average  $4.7 \times 10^4$  colony forming units (cfu) of *E. coli*, after a 2 to 5 second wipe and a 5 minute contact time this was reduced to an average of 7 cfu of *E. coli* per disc indicating a  $3.8 \log_{10}$  (99.99%) reduction in viable bacteria. The numbers of bacteria recovered from the wiped discs are so low that they would normally be considered statistically insignificant.

These results indicate that the Clinell wipes are highly effective in reducing the numbers of bacteria on steel surfaces. It is likely that the reduction in bacterial numbers is being constrained by the ability of *E. coli* to survive drying on a steel surface. If it were possible to achieve higher levels of loading it is likely that the Clinell wipes would achieve a greater reduction factor.

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## **Biocidal Surface Test - Clinell Wipes.**

### **1 Introduction**

The ability of Clinell wipes to disinfect contaminated steel surfaces was assessed using hospital grade stainless steel discs (9cm dia) pre-contaminated with an *E. coli* broth. These discs were air dried and then wiped once with a Clinell wipe, contact time between the wipe and the surface was between 2 and 5 seconds. Bacteria were recovered from the steel surfaces using pre-moistened swabs and enumerated via membrane filtration. The effectiveness of the wipes was assessed by comparison with un-wiped control discs contaminated in the same manner as the test discs.

### **2 Experimental Procedure**

#### ***2.1 Preparation of Test Discs***

Six 9 cm diameter discs of Hospital grade stainless steel were cleaned by soaking for >12 hours in 5% Decon 90 solution. These discs were then rinsed in distilled water and sterilized by soaking in 70% isopropyl alcohol for 12 hours. Excess isopropyl alcohol was poured off and the discs dried by evaporation in a Class II safety cabinet.

#### ***2.2 Preparation of the contamination fluid***

The contamination fluid consisted of a 24 hour culture of *E. coli* (ATCC 10536) grown at 37°C in Tryptone Soy broth (TSB). Prior to use the numbers of cfu/ml in the culture was determined via serial dilution in MRD<sup>1</sup> and duplicate pour plates using Tryptone Soy Agar (TSA). Plates were incubated at 37°C for 24 to 48 hours.

#### ***2.3 Contamination of Steel Discs***

Seven sterile, steel discs were placed in sterile Petri dishes and covered with 10ml of the contamination fluid. After a contact time of 5 minutes excess fluid was poured off and the discs were allowed to dry at 37°C in an incubator.

#### ***2.4 Analysis of Control Discs***

Three contaminated discs were chosen to act as controls. Each disc was swabbed with a sterile cotton wool swab pre moistened with MRD. After swabbing the swabs were placed in 10 mls of MRD and vortex mixed for 1 minute. Following mixing the cfu released by the swabs were determined via serial dilution in MRD and duplicate pour plates using TSA. Plates were incubated at 37°C for 24 to 48 hours.

#### ***2.5 Analysis of Contaminated Discs***

Three contaminated discs were wiped using a fresh Clinell wipe for each disc. The discs were wiped once for between 2 and 5 seconds and care was taken to ensure the entire disc was covered. The discs was then left for a contact time of five minutes and then swabbed with a sterile cotton wool swab pre

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<sup>1</sup> Maximum Recovery diluent supplied by LabM

moistened with MRD. After swabbing the swabs were placed in 10ml of neutralising solution for five minutes with occasional vortex mixing. After neutralisation the 10mls of neutraliser was filtered through a 0.45µm filter to recover any viable cells. The filters were placed on the surface of TSA plates and incubated for 24 to 48 hours at 37°C. The neutralizing solution employed consisted of 6g/l Tween 80, 6g/l Saponin, 0.2g/l l-histidine and 0.2g/l l-cysteine.

### 3 Results

Plate count results for the contamination fluid can be found in Table 1. These counts indicate that the contamination fluid had a total viable count of  $1.2 \times 10^9$  cfu/ml. After drying the average number of cfu per disc was  $4.7 \times 10^4$  (Table 2). This number fell to an average of 7 cfu per disc after wiping with the Clinell wipes (Table 3) giving a  $\log_{10}$  reduction in viable count of 3.8 and a percentage reduction of 99.99%.

Plate	Dilution				
	$10^{-5}$	$10^{-6}$	$10^{-7}$	$10^{-8}$	$10^{-9}$
1	TNC	TNC	122	19	3
2	TNC	TNC	112	18	1

Table 1. Contamination Fluid Total Viable Counts

Disk	Plate	Dilution	
		$10^{-2}$	$10^{-3}$
Disk 1	1	TNC	52
	2	TNC	43
Disk 2	1	TNC	66
	2	TNC	78
Disk 3	1	TNC	11
	2	TNC	30

Table 2. Control Disc Total Viable Counts

Volume Filtered (ml)	Disk		
	1	2	3
10.0	1	5	15

Table 3. Numbers of cfu Recovered from Wiped Discs

### 4 Summary and Conclusions

The aim of this work was to assess the effectiveness of Clinell biocidal wipes against bacteria dried onto a steel surface. The test is designed to simulate in use conditions and indicates that a single wipe lasting between 2 and 5 seconds produced a 3.8  $\log_{10}$  reduction in viable *E. coli* on the test surface. This equates to a 99.99% reduction in bacterial numbers. The number of bacteria recovered from the steel discs after treatment is very small and below what would be considered statistically significant. It is likely that the percentage and  $\log_{10}$  reductions reported here are constrained by how many viable bacteria are able to survive the drying process rather than the effectiveness of the wipes themselves. If it were possible to achieve higher levels of loading it is likely that the Clinell wipes would achieve a greater reduction factor.