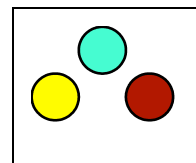


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Consulting Scientists to the Disinfectant Industry

24th September 2009

**Certificate of Analysis**

**Samples:** One sample of Zoonocide Z-71 received from Zoono Ltd, 20 Royston Court, Lichfield Road, RICHMOND, Surrey. TW9 3EH 15th September 2009.

**Certificate No:** 09J.056a.ZOO

**Page:** 1 of 2

**Sample Ref:** 9j / 056

**Analysis Required:** Adaptation of EN 13769 Quantitative Non Porous Surface Test for evaluation of bactericidal Activity of Chemical disinfectants.

**Samples Tested:** 22nd September 2008

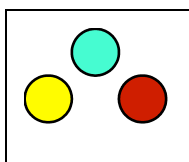
**Test Method.**

Twenty four glass slides were sprayed with Zoonocide, left to dry in a cool environment and then resprayed to give an even coating on the slide. These were again left to dry for several hours. Once dry the slides were rinsed with sterile deionised water and allowed to dry. These are the test slides. Similarly 24 glass slides were cleaned and sterilised with N - propanol and allowed to air dry. These are the control slides.

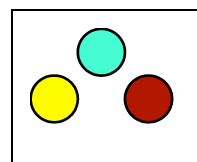
Six test slides and six control slides were each inoculated with 1ml of an overnight culture of the bacteria or a spore suspension of Aspergillus and spread to cover the whole of the surface and left at room temperature for a period of up to three hours. At time 0, 1 hour and 3 hours recovery of surviving organisms was carried out from two slides of each organism using 10ml of Nutrient broth and mechanical action to remove surface organisms. Serial dilutions were made from this suspension and plated out on to Tryptone soy Agar. The same procedure was carried out for control and test slides. Bacteria plates were incubated at 37°C for 48 hours and the Aspergillus at 30°C for 48 hours. Colony counts were recorded and the survival of each organism and each time interval calculated.

Identification of bacterial strain used - Pseudomonas aeruginosa ATCC 15442  
Escherichia coli NCTC 10418  
Staphylococcus aureus NCTC 10788  
Aspergillus niger NCTC 2275

D C Watson



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## Test Results

### *Pseudomonas aeruginosa*

Recovery Time	cfu/ml	log	log reduction	% Reduction
0 hours Control	$3.44 \times 10^4$	4.54		
Test	$3.36 \times 10^4$	4.53	0.01	N/A
1 hour Control	$2.08 \times 10^4$	4.32		
Test	$1.30 \times 10^2$	2.11	2.21	99.53
3 hours Control	$2.44 \times 10^4$	4.38		
Test	0	0	4.32	100

### *Escherichia coli*

Recovery Time	cfu/ml	log	log reduction	
0 hours Control	$2.96 \times 10^4$	4.47		
Test	$2.72 \times 10^4$	4.43	0.04	N/A
1 hour Control	$2.55 \times 10^4$	4.41		
Test	$5.0 \times 10^1$	1.70	2.71	99.80
3 hours Control	$2.36 \times 10^4$	4.37		
Test	0	0	4.37	100

### *Staphylococcus aureus*

Recovery Time	cfu/ml	log	log reduction	
0 hours Control	$5.60 \times 10^4$	4.75		
Test	$3.60 \times 10^4$	4.56	0.19	N/A
1 hour Control	$3.20 \times 10^4$	4.50		
Test	$2.08 \times 10^2$	2.32	2.18	99.35
3 hours Control	$2.41 \times 10^4$	4.38		
Test	0	0	4.38	100

### *Aspergillus niger*

Recovery Time	cfu/ml	log	log reduction	
0 hours Control	$4.84 \times 10^4$	4.68		
Test	$4.88 \times 10^4$	4.68	0	N/A
1 hour Control	$1.12 \times 10^4$	4.05		
Test	$2.30 \times 10^2$	2.36	1.69	97.95
3 hours Control	$1.38 \times 10^4$	4.14		
Test	$8.70 \times 10^1$	1.94	2.20	99.37