

CLIENT NAME: Keith MacGregor	CUSTOMER'S REF.: Ebiox REF: VAL-1
CLIENT ADDRESS : Ebiox Limited Whitehall Road Leeds LS1 4HR	
DRAFT	REPORT DATE: 19/12/02

## 1.0 **Aim**

To determine, against a variety of microorganisms commonly found in hospital environments, the bactericidal properties as a surface disinfectant of alcohol free wipes supplied by Ebiox.

## 2.0 **Materials and Equipment**

### 2.1 **Samples – Hard Surface Disinfectant Wipes**

2.1.1 Sample no. 12126175. Ebiox Bactericidal wipes – described as alcohol free medical grade disposable wet wipes containing organic hydroxyl, stabilizing agents, polymeric biguanide complex and surface-active agents.

### 2.2 **Test Organisms**

2.2.1 *Escherichia coli* ATCC 25922

2.2.2 *Staphylococcus aureus* ATCC 25923

2.2.3 *Bacillus subtilis* ATCC 6633

2.2.4 *Enterococcus faecalis* ATCC 29212

2.2.5 *Candida albicans* ATCC 2091

2.2.6 *Methicillin resistant Staphylococcus aureus* (MRSA) NCTC 10442

2.2.7 *Pseudomonas aeruginosa* ATCC 27853

2.2.8 *Streptococcus pyogenes* ATCC 19615

2.2.9 *Proteus mirabilis* ATCC 14153

### 2.3 **Media**

2.3.1 Tryptone Soya Broth (TSB) – Oxoid CM 129 – a highly nutritious medium used to support the growth of the test organisms.

2.3.2 Maximum Recovery Diluent (MRD) – LAB M LAB 103 – an osmotically controlled diluent used to dilute the test organisms after overnight growth in TSB. This formula is recommended by ISO 6887:BS 5763.

2.3.3 Maximum Recovery Diluent with 1% Bovine Albumin (MRDBA) – as 2.3.2 but with the addition of 1% bovine albumin which mimics soiling or "dirty conditions" as recommended in BSI DD 177:1988 (confirmed August 1993) – Method of test for the antimicrobial activity of disinfectants in food hygiene.

2.3.4 Neutralising Solution – Q Laboratories' Media Recipe No.28 – a neutralizing buffer with the ability to inactivate the effect of disinfectant agents.

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2.3.5 Plate Count Agar – LAB M LAB 149 – this medium is used to establish total viable counts for aerobic organisms and is formulated to A.P.H.A. specification.

## 2.4 Equipment

2.4.1 Petri Dishes – 90mm

2.4.2 Incubator 30°C ± 1°C

2.4.3 Pipettes – 250 µl and 1 ml capacity

2.4.4 L-shaped sterile spreaders

## 3.0 Test Method

3.1 Each of the test organisms was inoculated into a Tryptone Soya Broth and incubated overnight at 30°C ± 1°C

3.2 A large metal surface, similar to hospital surfaces, was divided into areas of 5cm square, disinfected using alcohol and allowed to air dry.

3.3 Each of the overnight broths of the test organisms were diluted 1:1000 in MRD to represent clean surface conditions and in MRDBA to represent 'dirty' surface conditions.

3.4 250 µl of each of the diluted test organisms in MRD was added to four of the designated disinfected squares per organism, spread with L-shaped spreaders and allowed to air dry.

3.5 Section 3.4 was repeated but using the diluted test organisms in MRDBA. See Appendix 1 for the template.

3.6 One of the squares was wiped with a test wipe, left for one minute then swabbed with a cotton tipped swab over the full surface of the square. The swab was then placed into 9ml neutralizing solution (-dilution) and diluted 10 fold from -2 to -3 dilutions. 1ml from each of the -1, -2 and -3 dilutions was added to sterile petri dishes. Molten Plate Count Agar was added to the plates and mixed well.

3.7 Section 3.6 was repeated but an inoculated square was wiped with a fresh wipe and left for 2 minutes, another square left for 5 minutes before swabbing and another square swabbed without any wipes being used. This was the control square.

3.8 Sections 3.6 and 3.7 was repeated for each of the organisms in MRD and MRDBA with each of the 2 test samples of alcohol wipes.

3.9 All the petri dishes containing Plate Count Agar were allowed to dry, inverted and incubated at 30°C + 1°C for 3 days, based on BSI 5763 Part 1:1991. The colonies were then counted and the number of colony forming units (cfu) per swab calculated.

## 4.0 Results

4.1 See Tables for numbers of organisms left on the test squares after wipe treatment and before wipe treatments (control square).

4.2 The wipes significantly reduced the numbers of test organisms to <10 cfu/swab after a contact time of 1 minute.

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## 5.0 Conclusion

- 5.1 The wipes were effective as a disinfectant against the 9 microorganisms tested, even in dirty conditions
- 5.2 A contact time of 1 minute of the wipes tested resulted in a significant reduction in organism numbers.
- 5.3 This Ebiox wipes were effective, in less than 1 minute, in reducing the 9 microorganisms to a threshold below which it was not possible to accurately measure their viability. This result is at a minimum, comparable with good quality alcohol disinfectant wipes. The Ebiox wipes are alcohol free and carry no flammability hazard warning.

SIGNED: .....

NAME: NICOLETTE O'REILLY      SUSAN CLAY      SHIRLEY GORDON

POSITION: MICROBIOLOGIST      GROUP QUALITY MANAGER      LABORATORY MANAGER

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WIPE EVALUATION												
SAMPLES DETAILS:- Ebiox Wipes Bactericidal wipes												
ORGANISMS	Number of organisms (cfu/swab) remaining after treatment with Ebiox alcohol free wipes under clean conditions					Number of organisms (cfu/swab) remaining after treatment with Ebiox alcohol free wipes under dirty conditions						
	CONTROL	1 MINUTE	3 MINUTES	5 MINUTES	CONTROL	1 MINUTE	3 MINUTES	5 MINUTES	CONTROL	1 MINUTE	3 MINUTES	5 MINUTES
E. COLI	1100	<10	<10	<10	1500	<10	<10	<10	1500	<10	<10	<10
STAPHYLOCOCCUS AUREUS	2600	<10	<10	<10	3900	<10	<10	<10	3900	<10	<10	<10
BACILLUS SUBTILIS	3100	<10	<10	<10	7100	<10	<10	<10	7100	<10	<10	<10
ENTEROCOCCUS FAECALIS	3900	<10	<10	<10	7200	<10	<10	<10	7200	<10	<10	<10
CANDIDA ALBICANS	420	<10	<10	<10	1200	<10	<10	<10	1200	<10	<10	<10
MRSA	510	<10	<10	<10	940	<10	<10	<10	940	<10	<10	<10
PSEUDOMONAS AERUGINOSA	3200	<10	<10	<10	11000	<10	<10	<10	11000	<10	<10	<10
STREPTOCOCCUS PYOGENES	2100	<10	<10	<10	11000	<10	<10	<10	11000	<10	<10	<10
PROTEUS MIRABILIS	3200	<10	<10	<10	1100	<10	<10	<10	1100	<10	<10	<10