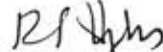
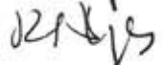


**Bactericidal activity of GAMA Healthcare Ltd.
biocide determined using the European Standard
Test method BS EN 1276:1997 against: *Salmonella
typhimurium* ATCC 14028.**

July 2006

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Signature: 
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Date: 25/7/06
Date: 25/7/06



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Tests Carried Out By:

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Microbiological Tests

Test Method British/European Standard BS EN 1276:1997.
Dilution-neutralisation

Test Procedures Full details of all the test and control procedures
used are given in the Test Method

Disinfectant GAMA Healthcare Ltd biocide
Batch number: N/A
Date of delivery: June 2006
Storage conditions: 20°C – 25°C
Active substances: not specified
Appearance product dilutions: colourless, clear
product solution.

Interfering Substance (Organic Challenge)

1. Simulated clean conditions:
0.3 g l⁻¹ bovine albumin (final
concentration)
2. Simulated dirty conditions:
3.0 g l⁻¹ bovine albumin (final
concentration)

Temperature Ambient (25°C)

Contact Time Tested 5 (± 10 s) minute.

Test Organisms *Salmonella typhimurium* ATCC 14028

Culture Medium Tryptone Soya Agar, Lab M

Incubation Plates were incubated at 37 °C for 24 - 48 h

Diluent MRD, Lab M

Neutraliser Neutraliser, containing 60g/l Tween 80, 60g/l
Saponin, 2g/l L-histidine, 2g/l L-cysteine in
MRD.

General Method

A standard suspension of test organisms containing $1.5 - 5.0 \times 10^8$ cells ml^{-1} of bacteria was prepared. 1 ml of interfering substance was pipetted into a Universal bottle, followed by 1 ml of test organism suspension. The mixture was mixed and left for 2 minutes. After 2 minutes 8 ml of disinfectant was added and mixed. In this case the disinfectant was the GAMA Healthcare Ltd biocide. After a contact time of 5 minutes, a 1 ml sample of the reaction mixture was pipetted into 9 ml of neutraliser and left for 5 minutes. A 1 ml sample was then pipetted into 2 Petri dishes and mixed with 15 ml of culture medium tempered at 47 °C. After setting, the Petri dishes were incubated at 37 °C. Colony forming units were counted after 1-2 days incubation and the fraction of surviving organisms calculated.

Requirements of this standard

The product, when tested as stipulated under simulated clean conditions (0.3 g l^{-1} bovine albumin) or dirty conditions (3 g l^{-1} bovine albumin) under the required test conditions (25°C, 5 minute contact, for the selected reference strain), shall demonstrate at least a 5 \log_{10} reduction in viable counts.

Results¹

Results from the test are summarised in Tables 1 and 2, a full set of results can be found in Table 3.

Test Conditions	Contact Time (minutes)	Log ₁₀ Reduction Achieved
0.3 g l^{-1} (clean)	5	>5 ¹
3.0 g l^{-1} (dirty)	5	>5 ¹

Table 1. Log₁₀ reductions in *S. typhimurium* viable counts following a 5 minute exposure to the test material.

Referenced Organism	Starting concentration CFU ml^{-1}	Final concentration CFU ml^{-1} clean 0.3 g l^{-1} Bovine Albumin	Final concentration CFU ml^{-1} dirty 3.0 g l^{-1} Bovine Albumin
<i>Salmonella typhimurium</i> ATCC14028	1.7×10^8 (171,160 ¹ , 20, 22 ²)	Plate count 3, 3. (Actual 6 \log_{10} reduction)	Plate count 5, 2. (Actual 6 \log_{10} reduction)
CFU = colony forming units ¹ viable count of bacterial colonies, 1 ml sample of 10^{-6} bacterial suspension ² viable count of bacterial colonies, 1 ml sample of 10^{-7} bacterial suspension			

Table 2. Reductions in *S. typhimurium* viable counts following a 5 minute exposure to the test material.

¹ See Table of results in Appendix 1.

Interpretation of the Results

When tested against *S. typhimurium* (ATCC 14028) with a 5 minute contact time a full strength GAMA Healthcare Ltd biocide met the requirements of the Standard at ambient temperature (25°C) under simulated clean and dirty conditions.

Conclusion

According to EN 1276:1997, the batch provided of GAMA Healthcare Ltd biocide possesses bactericidal activity in 5 minutes at ambient temperature (25°C) under clean conditions (0.3g/l bovine albumin) and dirty conditions (3g/l bovine albumin) for referenced strain *S. typhimurium* (ATCC 14028).

Signed:

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