

TITLEBACTERICIDAL ACTIVITY OF ENDUROCID E UNDER
DIRTY CONDITIONSCONTACT AT
TESTING LABORATORY

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DD10 0SPIDENTIFICATION OF SAMPLE

Name of sample: ENDUROCID E

Laboratory Number: 02/10073

Batch Number: OTL/SS 01/07.03

Appearance: Clear liquid

Storage conditions: Room Temperature

Date of receipt: 10/07/03

Date(s) of test(s): 30/07/03 – 06/08/03

TEST METHODpr EN 13713:1999 Chemical disinfectants and antiseptics – Surface
disinfectants used in human medicine, bactericidal activity – Test
method and requirements.

EXPERIMENTAL CONDITIONS

| | |
|----------------------------|--|
| Procedure: | Membrane filtration method used as a suitable neutralizer was not Found. (See report number BIO/04073) |
| Test dilutions: | As received i.e. undiluted It is not possible to determine the bactericidal activity of an undiluted product as some dilution is always produced by adding the inoculum and interfering substance. Ready to use products are therefore actually tested at a concentration of 80%. |
| <u>DEVIATION:</u> | The sample was only tested as received three dilutions were not tested as specified in the standard |
| Test temperature: | 20°C +/- 1°C |
| Test conditions: | Dirty:- conditions representative of surfaces which are known to or may contain, organic and/or inorganic materials. (final concentration of yeast extract 5g/l and bovine albumin 5g/l) |
| Contact time: | 5 minutes |
| Temperature of incubation: | 37°C +/- 1°C |
| Test organisms: | <i>Enterococcus hirae</i> NCIMB 8191 <i>Escherichia coli</i> NCTC 10418 <i>Pseudomonas aeruginosa</i> NCIMB 10421 <i>Staphylococcus aureus</i> NCTC 10788 |

REQUIREMENTS

The product when diluted in hard water and tested in accordance with clause 5* prEN 13713, under the required test conditions (20°C, 5 min, four referenced strains), shall demonstrate at least a 10⁵ log reduction in viable counts.

*The validation test (see Annex A prEN 13713) shall also be carried out at the same time as the test procedure using only the highest product concentration and the same conditions as used in the test.

A product which passes the test, is characterized as a chemical possessing bactericidal activity under conditions representative of practical use. Further laboratory tests using surfaces simulating practical conditions are required to qualify the product as a surface disinfectant.

RESULTS VALIDATION TESTSTABLE 2 Verification of the methodology and validation of membrane filtration method for ENDUROCID under dirty conditions

| Organism | Viable count (cfu/ml) | | | | |
|-------------------------------|-------------------------------|--|---|------------------------|-----------------------------|
| | Bacterial test suspension (N) | Bacterial suspension (N _v) | Validation of experimental conditions (A) | Filtration control (B) | Filtration test control (C) |
| <i>Enterococcus hirae</i> | 2.8 x 10 ⁸ | 1.2 x 10 ³ | 9.8 x 10 ¹ | 9.3 x 10 ¹ | 7.7 x 10 ¹ |
| <i>Escherichia coli</i> | 2.2 x 10 ⁸ | 8.8 x 10 ³ | 6.8 x 10 ¹ | 9.2 x 10 ¹ | 5.0 x 10 ¹ |
| <i>Pseudomonas aeruginosa</i> | 2.6 x 10 ⁸ | 1.5 x 10 ³ | 7.6 x 10 ¹ | 8.0 x 10 ¹ | 7.6x 10 ¹ |
| <i>Staphylococcus aureus</i> | 4.3 x 10 ⁸ | 9.6 x 10 ³ | 6.8 x 10 ¹ | 7.4 x 10 ¹ | 7.2 x 10 ¹ |

KEY:

For the strain tested:

N is between 1.5 x 10⁸ colony forming units per millilitre (cfu/ml) and 5 x 10⁸ cfu/mlN_v is between 6 x 10² cfu/ml and 3 x 10³ cfu/mlA is equal to or greater than 0.05 x N_vB is equal to or greater than 0.05 x N_v

C is equal to or greater than 0.5 x B

NOTE: The test method is validated for the referenced strains

RESULTS TEST METHODTABLE 2 Reduction in viability of *Enterococcus hirae* by ENDUROCID E under dirty conditions at 20°C with a 5 minute contact time

| Dilution | Viable counts for the test mixture | Viable counts (cfu/ml) for the test mixture (N _a) | Reduction in viability (N x 10 ⁻¹ ÷ N _a) | Status |
|-----------|------------------------------------|---|---|--------|
| Undiluted | 0, 0 | <1.5 x 10 ² | >1.9 x 10 ⁵ | PASS |

TABLE 3 Reduction in viability of *Escherichia coli* by ENDUROCID E under dirty conditions at 20°C with a 5 minute contact time

| Dilution | Viable counts for the test mixture | Viable counts (cfu/ml) for the test mixture (N _a) | Reduction in viability (N x 10 ⁻¹ ÷ N _a) | Status |
|-----------|------------------------------------|---|---|--------|
| Undiluted | 0, 1 | <1.5x 10 ² | >1.5 x 10 ⁵ | PASS |

KEY:

- > = Greater than.
 < = Less than
 PASS = ≥ 10⁵ reduction in viability
 FAIL = < 10⁵ reduction in viability

RESULTSTEST METHOD

TABLE 4 Reduction in viability of *Pseudomonas aeruginosa* by ENDUROCID E under dirty conditions at 20°C with a 5 minute contact time

| Dilution | Viable counts for the test mixture | Viable counts (cfu/ml) for the test mixture (N_a) | Reduction in viability ($N \times 10^{-1} \div N_a$) | Status |
|-----------|------------------------------------|---|--|--------|
| Undiluted | 0, 0 | $<1.5 \times 10^2$ | $>1.7 \times 10^5$ | PASS |

TABLE 5 Reduction in viability of *Staphylococcus aureus* by ENDUROCID E under dirty conditions at 20°C with a 5 minute contact time

| Dilution | Viable counts for the test mixture | Viable counts (cfu/ml) for the test mixture (N_a) | Reduction in viability ($N \times 10^{-1} \div N_a$) | Status |
|-----------|------------------------------------|---|--|--------|
| Undiluted | 0, 0 | $<1.5 \times 10^2$ | $>2.9 \times 10^5$ | PASS |

KEY:

> = Greater than.

< = Less than

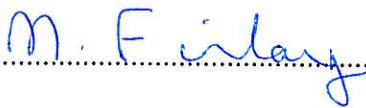
PASS = $\geq 10^5$ reduction in viability

FAIL = $< 10^5$ reduction in viability

CONCLUSION

According to prEN 13713:1999, the sample of ENDUROCIDe batch number OTL/SS 01/07.03, when used undiluted possesses bactericidal activity in five minutes at 20°C under dirty conditions for the referenced strains *Enterococcus hirae*, *Escherichia coli*, *Pseudomonas aeruginosa* and *Staphylococcus aureus*.

The product when used undiluted is suggested as being suitable for practical situations of use.

SIGNATURE.....
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