

DETERMINATION OF FUNGICIDAL OR YEASTICIDAL ACTIVITY FOR SURFACE DISINFECTION

ACCORDING TO UNE EN 13624:2013

TEST REPORT N° 01/2023

| | |
|---|---|
| a) Identification of the test laboratory | Laboratório de Microbiologia Aplicado à Saúde (LMAS) Centro de Engenharia Biológica, Universidade do Minho |
| b) Identification of the client | |
| - Name | Success Gadget, Nanotecnologia e Novos Materiais, Lda |
| - Address | Rua Filipa Borges, 1245, 4750-823 Barcelos, Portugal |
| c) Identification of the sample (data provided by the client, the laboratory is not responsible for the information provided by the client) | |
| - Product name | Care Us |
| - Batch number | AMS220707 |
| - Expiration date | 2023.07 |
| - Storage conditions | Ambient temperature |
| - Manufacturer | Success Gadget, Nanotecnologia e Novos Materiais, Lda |
| - Manufacturer recommended product diluent | Not disclosed |
| - Active substances and their concentration | Hydrogen Peroxide (1.4% (w/w)); Ethanol (50% (w/w)) |
| - Product Appearance | Whitish (Please shake before use) |
| d) Description of the sample by the laboratory | |
| - Delivery Date | 13/07/2022 |
| - Product Appearance | Transparent Liquid |
| - Active substances and their concentration | Not request |
| e) Test method and its validation | |
| - Method | Dilution - neutralization |
| - Technique | Pour plate |
| - Neutralizer | Lecithin (3g/l); Tween 80 (30ml/l); Saponin (30 g/l); L-hystidine (1g/l); Sodium thiosulfate(5g/l); In phosphate buffer 0,0025N |
| f) Experimental Conditions | |
| - Period of analysis | From 18/07/2022 to 29/07/2022 |
| - Diluent of the product used during the test | Sterile distilled water |
| - Product test concentration | 80%; 40%; 20% |
| - Aspect of product dilution | Transparent solutions, soluble in sterile distilled water |
| - Stability of the mixture during the procedure | Stable |
| - Interfering substances | Aqueous bovine albumin solution 0.3 g/L |
| - Contact times | 60 seconds ± 5 seconds |
| - Temperature of assay | (20 ± 1) °C |
| - Temperature of incubation | (30 ± 1) °C |
| - Identification of the test strains | - <i>Candida albicans</i> SC 5314 - <i>Aspergillus niger</i> MUM 9235 |
| g) Test results (See table A) | |
| h) Special remarks regarding the results | |
| - All controls and validation were within their baseline limits. | |
| - At least one concentration of the product demonstrated a log reduction of less than 4 lg. | |
| - No precipitate was formed during the test procedure (test mixtures were homogeneous). | |
| i) Conclusion | |
| For the tested sample of the product "Care Us" batch AMS220707, UNDILUTED (80%) shows yeasticidal and fungicidal activity, based on EN 13624:2013 under clean conditions (aqueous bovine albumin solution 0.3 g/L) at 20 °C, for 60 second of contact. | |

Technical manager

Laboratory technician

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| Test Organisms | Validation test suspension | | | | Assay validation | | | | | | Assay Suspension | | | Test procedure at the concentration, % | | |
|--------------------------------------|--|----------|---------------|----------|-------------------------------------|----------|-------------------------|----------|-----------------------|----------|------------------|--|--|--|--|--|
| | N_V and N_{V0} | | N_{VB} | | Experimental conditions control (A) | | Neutralizer control (B) | | Method validation (C) | | | | | 80 | 40 | 20 |
| <i>Candida albicans</i> SC 5314 | V_{C1} | V_{C2} | V_{C1} | V_{C2} | V_{C1} | V_{C2} | V_{C1} | V_{C2} | V_{C1} | V_{C2} | 10^{-5} | V_{C1} | V_{C2} | $10^0:0; 0$ $10^{-1}:0; 0$ $N_a = < 1.4 \times 10^2$ $\text{Log } N_a = < 2.15$ $\text{Log } R = > 4.23$ | $10^0: >330; >330$ $10^{-1}: >330; >330$ $N_a = > 3.3 \times 10^4$ $\text{Log } N_a = > 4.52$ $\text{Log } R = < 1.86$ | $10^0: >330; >330$ $10^{-1}: >330; >330$ $N_a = > 3.3 \times 10^4$ $\text{Log } N_a = > 4.52$ $\text{Log } R = < 1.86$ |
| | 41 | 48 | 34 | 32 | 61 | 99 | 39 | 38 | 50 | 46 | | 238 | 236 | | | |
| | $N_V = 4.4 \times 10^2$ $N_{V0} = 44$ | | $N_{VB} = 44$ | | A = 80 | | B = 38 | | C = 48 | | 10^{-6} | 26 | 24 | | | |
| | | | | | | | | | | | | $N = 2.4 \times 10^7$ $\text{Log } N = 7.38$ | $N_0 = 2.4 \times 10^6$ $\text{Log } N_0 = 6.38$ | | | |
| <i>Aspergillus niger</i> MUM 9235 | V_{C1} | V_{C2} | V_{C1} | V_{C2} | V_{C1} | V_{C2} | V_{C1} | V_{C2} | V_{C1} | V_{C2} | 10^{-5} | V_{C1} | V_{C2} | $10^0:0; 9$ $10^{-1}:0; 0$ $N_a = < 1.4 \times 10^2$ $\text{Log } N_a = < 2.15$ $\text{Log } R = > 4.06$ | $10^0: >330; >330$ $10^{-1}: >330; >330$ $N_a = > 3.3 \times 10^4$ $\text{Log } N_a = > 4.52$ $\text{Log } R = < 1.69$ | $10^0: >330; >330$ $10^{-1}: >330; >330$ $N_a = > 3.3 \times 10^4$ $\text{Log } N_a = > 4.52$ $\text{Log } R = < 1.69$ |
| | 49 | 51 | 42 | 44 | 50 | 49 | 88 | 72 | 53 | 53 | | 175 | 144 | | | |
| | $N_V = 5.0 \times 10^2$ $N_{V0} = 50$ | | $N_{VB} = 43$ | | A = 50 | | B = 80 | | C = 53 | | 10^{-6} | 18 | 16 | | | |
| | | | | | | | | | | | | $N = 1.61 \times 10^7$ $\text{Log } N = 7.21$ | $N_0 = 1.61 \times 10^6$ $\text{Log } N_0 = 6.21$ | | | |

V_c: counts obtained in plate.

N: number of cells per ml in the test suspension.

N₀: number of cells per ml at the beginning of the contact time.

N_v: number of cells per ml in the validation suspension.

N_{v0}: number of cells per ml in mixtures A, B and C at the beginning of the contact time.

N_{VB}: (dilution method) number of cells per ml after 1: 100 dilution

N_a: number of survivors per ml in the test mixture at the end of the contact time.

A: number of survivors in the control conditions of experiment A.

B: number of survivors in the neutralizing or filtrate control B.

C: number of survivors in the validation of method C.

Log R: Logarithmic reduction.

Verification of the methodology:

- N** is comprised between 1.5 and 5×10^7 ($7.17 < \log N < 7.70$).
N (modified method) is in the range of 1.5 to 5×10^8 ($8.17 < \log N < 8.70$).
N₀ is between 1.5 and 5×10^6 ($6.17 < \log N < 6.70$).
- N_{v0}** is between 30 and 160.
N_v is between 3×10^2 and 1.6×10^3 .
N_v (modified method) is between 3×10^3 and 1.6×10^4 .
N_{VB} is between 3×10^4 and 1.6×10^5 .
- A, B and C** are equal to or greater than $0.5 \times N_{v0}$
B (dilution-neutralization) is equal to or greater than $0.0005 \times N_{VB}$
- For the results calculated by weighted averages of two subsequent dilutions, the quotient of the mean of the 2 results is between 5 and 15