

CERTIFICATE OF ANALYSIS

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EXCELSIA TECHNOLOGIES SDN BHD
UNIT 103, 1ST FLOOR, LIF NO.2, BLOCK C,
DAMANSARA INTAN, NO.1, JALAN SS20/27, 47400
PETALING JAYA, SELANGOR.

LAB NO. : CAW/1903/82766
SAMPLE ID : CAW/1903/82766/227399
SAMPLE MARKING : ULTRA MIST HERBAL SOLUTION
RECEIVED DATE : 22ND MARCH 2019
REPORTED DATE : 29TH MARCH 2019

Test: Challenge Test

Organisms used to challenge:

- a) *Staphylococcus aureus* (ATCC 6538)
- b) *Escherichia coli* (ATCC 8739)
- c) *Pseudomonas aeruginosa* (ATCC 9027)
- d) *Candida albicans* (ATCC 10231)
- e) *Aspergillus brasiliensis* (ATCC 16404)

Procedure:

- 1) Subculture the microorganisms from pure working culture onto non-selective media as follow:
 - a) *S. aureus*, *P. aeruginosa* and *E. coli* on Tryptic Soy Agar
 - b) *C. albicans* and *A. brasiliensis* on Sabouraud Dextrose agar
- 2) Incubate *S. aureus*, *P. aeruginosa* and *E. coli* at 35°C for 24 hours, while incubate *C. albicans* and *A. brasiliensis* at 25°C for 3-5 days
- 3) Prepare inoculum suspension to be spiked into sample
- 4) Dilute and plate out the inoculum onto their respective media to determine microbial count of inoculum suspension.
- 5) Spread plate 0.1ml of 10⁷ cfu/ml inoculum suspension onto an even surface
- 6) Spray the sample on the surface and let dry
- 7) Do swab sampling at each contact time. Then, carry out serial dilution using diluent containing 8% Polysorbate 80 for Bactashield Ultimate. Polysorbate 80 acts as neutralizer to inhibit the disinfectant property of the sample, so actual efficacy of sample can be studied for specific contact time.
- 8) Transfer 1ml of each dilution on petri dish and perform pour plate with respective media. Incubate the plates with condition as follow:
 - a) *S. aureus*, *P. aeruginosa* and *E. coli* - 35°C for 72h
 - b) *C. albicans* and *A. brasiliensis* - 25°C for 3-5d

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*Initial

Test Parameter	Inoculum Used (cfu/ml)	Inoculum Recovered - 0min	**Percentage Killed (%)
<i>S. aureus</i>	1.5×10^6	3.1×10^4	97.933%
<i>E. coli</i>	1.7×10^6	2.8×10^4	98.353%
<i>P. aeruginosa</i>	1.2×10^6	2.7×10^4	97.750%
<i>C. albicans</i>	4.8×10^5	5.7×10^3	98.881%
<i>A. brasiliensis</i>	3.3×10^5	6.2×10^3	98.121%

*Contact time at 5 minute

Test Parameter	Inoculum Used (cfu/ml)	Inoculum Recovered - 5mins	**Percentage Killed (%)
<i>S. aureus</i>	1.5×10^6	6.4×10^3	99.573%
<i>E. coli</i>	1.7×10^6	3.4×10^3	99.800%
<i>P. aeruginosa</i>	1.5×10^6	3.5×10^3	99.767%
<i>C. albicans</i>	4.8×10^5	7.5×10^2	99.844%
<i>A. brasiliensis</i>	3.3×10^5	6.7×10^2	99.797%

*Contact time at 15 minute

Test Parameter	Inoculum Used (cfu/ml)	Inoculum Recovered - 15mins	**Percentage Killed (%)
<i>S. aureus</i>	1.5×10^6	1.1×10^2	99.993%
<i>E. coli</i>	1.7×10^6	2.0×10^2	99.988%
<i>P. aeruginosa</i>	1.2×10^6	3.0×10^2	99.975%
<i>C. albicans</i>	4.8×10^5	3.5×10^2	99.927%
<i>A. brasiliensis</i>	3.3×10^5	1.4×10^2	99.958%

*Contact time at 30 minute

Test Parameter	Inoculum Used (cfu/ml)	Inoculum Recovered - 30mins	**Percentage Killed (%)
<i>S. aureus</i>	1.5×10^6	2.2×10^1	99.999%
<i>E. coli</i>	1.7×10^6	1.3×10^1	99.999%
<i>P. aeruginosa</i>	1.2×10^6	3.0×10^1	99.998%
<i>C. albicans</i>	4.8×10^5	1.6×10^1	99.997%
<i>A. brasiliensis</i>	3.3×10^5	2.1×10^1	99.994%

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*Contact time at 60 minute

Test Parameter	Inoculum Used (cfu/ml)	Inoculum Recovered - 60mins	**Percentage Killed (%)
<i>S. aureus</i>	1.5×10^6	NG <10	99.999%
<i>E. coli</i>	1.7×10^6	NG <10	99.999%
<i>P. aeruginosa</i>	1.2×10^6	NG <10	99.999%
<i>C. albicans</i>	4.8×10^5	NG <10	99.998%
<i>A. brasiliensis</i>	3.3×10^5	NG <10	99.997%

*Contact time at 3 hours

Test Parameter	Inoculum Used (cfu/ml)	Inoculum Recovered - 3hours	**Percentage Killed (%)
<i>S. aureus</i>	1.5×10^6	NG <10	99.999%
<i>E. coli</i>	1.7×10^6	NG <10	99.999%
<i>P. aeruginosa</i>	1.2×10^6	NG <10	99.999%
<i>C. albicans</i>	4.8×10^5	NG <10	99.998%
<i>A. brasiliensis</i>	3.3×10^5	NG <10	99.997%

* Contact time is time of contact between sample and culture

** Percentage Killed = $\frac{\text{Inoculum used} - \text{Inoculum Recovered}}{\text{Inoculum Used}} \times 100\%$

Note:

- 1) NG means No Growth
- 2) cfu means Colony Forming Unit
- 3) Opinion and Interpretation expressed herein are outside the scope of SAMM accreditation. The above results relate only to the items tested. This report shall not be reproduced, without the written approval of Chemsil Air & Water Sdn. Bhd.

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