QACS LAB
QUALITY ASSURANCE & CONTROL SYSTEMS

A: 1 Antigonis str, 14451, Athens, Greece
T: (+30) 210 2934745 | E: info@qacs.gr
F: (+30) 210 2934606 | www.qacslab.com

STUDY REPORT

2021-1701/21 23 00161

Active chlorine released from hypochlorous acid by electrochemical by activation

SUSPENSION TEST
ACCORDING TO EN 1650:2019
(Phase 2 step 1)

Chemical disinfectants and antiseptics
Quantitative suspension test for the evaluation of fungicidal or yeasticidal activity of chemical disinfectants and antiseptics used in food, industrial, domestic and institutional areas - Test method and requirements (phase 2, step 1)

MARCH 2021

QACS LTD, 1 Antigonis str, 144-51 Metamorfossi, Greece. Tel: +30-210-2934745, fax: + 30-210-2934606, email: info@qacs.gr website: www.qacslab.com

STUDY REPORT 2021-1701/21 23 00161

SUSPENSION TEST ACCORDING TO EN 1650:2019
Chemical disinfectants and antiseptics – evaluation of fungicidal or yeasticidal activity of chemical disinfectants and antiseptics used in food, industrial, domestic and institutional areas – Test method and requirements (Phase 2 step 1)

A: 1 Antigonis str, 14451, Athens, Greece
T: (+30) 210 2934745 | E: info@qacs.gr
F: (+30) 210 2934606 | www.qacslab.com

TEST PRODUCT IDENTIFICATION

SCOPE

This document specifies a test method and the minimum requirements for yeasticidal or fungicidal activity of chemical disinfectant and antiseptic products that form a homogeneous, physically stable preparation when diluted with hard water or - in the case of ready-to-use-products - with

PRODUCT NAME	:	Active chlorine released from hypochlorous acid by electrochemical by activation
ACTIVE SUBSTANCES	:	Active chlorine released from hypochlorous acid 0,05%
APPEARANCE OF THE PRODUCT	:	Liquid
STORAGE CONDITIONS	:	Room temperature, darkness
TEST CONDITIONS	:	Test conducted at 20ºC □ 1 ºC
LOT	:	02 – bacteria 1
METHOD	:	EN 1650:2019
RECEIPT DATE	:	17/02/2021
STUDY PERIOD	:	03/03/2021-05/03/2021
LAB ID	:	2021-1701/21 23 00161

water. Products can only be tested at a concentration of 80 % or less as some dilution is always produced by adding the test organisms and interfering substance.
This document applies to products that are used in food, industrial, domestic and institutional areas excluding areas and situations where disinfection is medically indicated and excluding products used on living tissues except those for hand hygiene in the above considered areas.

PRINCIPLE

A sample of the product as delivered and/or diluted with hard water (or water for ready-to-use products) is added to a test suspension of fungi (yeast cells or mould spores) in a solution of an interfering substance. The mixture is maintained at the chosen test temperature for the adopted contact time. At the end of this contact time, an aliquot is taken, and the yeasticidal and/or fungicidal activity in this portion is immediately neutralized or suppressed by a validated method. The method of choice is dilution-neutralization. If a suitable neutralizer cannot be found, membrane filtration is used. The numbers of surviving yeasts in each sample are determined and the reduction is calculated.
The test is performed using only the vegetative cells of *Candida albicans* (yeasticidal activity) and the spores of *Aspergillus brasiliensis* (fungicidal activity) as test organisms.

TEST CONDITIONS

1. The following procedure was performed in water bath at 20 °C.
2. The test product was tested at 15 seconds contact time.
3. Interfering substance: A final concentration of 0.3g/L bovine albumin was used for testing (clean conditions).
4. Neutralization Method used: Dilution neutralization.
5. Neutralizer used: LPT Dilution Broth containing polysorbate 80.
6. According to EN 1650, products shall be tested at a minimum of three different concentrations to include one concentration in the active range and one concentration in the non-active range. In this case the product was tested: Undiluted (80%), 50%, 1%.

A: 1 Antigonis str, 14451, Athens, Greece
T: (+30) 210 2934745 | E: info@qacs.gr
F: (+30) 210 2934606 | www.qacslab.com

TEST MICROORGANISMS

Candida albicans	ATCC 10231
Aspergillus brasiliensis	ATCC 16404

FUNGICIDAL ACTIVITY FOR GENERAL PURPOSES

The product shall be deemed to have passed the EN 1650 standard if it demonstrates in a valid test a reduction of at least a 4 lg under the adopted test conditions with the chosen interfering substance simulating clean or dirty conditions defined by this European Standard when the test organisms are Candida albicans and Aspergillus brasiliensis.

YEASTICIDAL ACTIVITY FOR GENERAL PURPOSES

The product shall be deemed to have passed the EN 1650 standard (yeasticidal activity) if it demonstrates in a valid test a reduction of at least a 4 lg within the adopted test conditions with the chosen interfering substance simulating clean or dirty conditions defined by this European Standard when the test organism is Candida albicans.

ASSAY ACCEPTANCE CRITERIA

1. Test Suspension (N) is between 1.5 to 5.0 X 10^7 CFU per mL (7.17≤ log No ≤7.70).
2. No (N/10) is between 1.5 to 5.0 X 10^6 CFU per mL (6.17 ≤ log No ≤ 6.70).
3. Validation Suspension=Nv is between 3.0 x 10^2 and 1.6 x 10^3.
4. Nvo (Nv/10) is between 30 and 160.
5. Na is the number of survivors (cells) per ml in the test mixture at the end of contact time.
6. R (log reduction) = No - Na
7. Average recovery values for the experimental conditions control (A) were equal to or greater than 0.5 times the Validation Suspension (Nvo)
8. Average recovery values for the Neutralizer control (B) were equal to or greater than 0.5 times the Validation Suspension (Nvo)
9. Average recovery values for the Method Validation control (C) were equal to or greater than 0.5 times the Validation Suspension (Nvo)
10. Control of weighted mean counts. Quotient is not lower than 5 and not higher than 15

ARCHIVING

The laboratory book which contains all the information (raw data and results) regarding the study and the study reports are kept in the laboratory archives for 5 years.

QACS LAB
QUALITY ASSURANCE & CONTROL SYSTEMS

A: 1 Antigonis str, 14451, Athens, Greece
T: (+30) 210 2934745 | E: info@qacs.gr
F: (+30) 210 2934606 | www.qacslab.com

Test Results for Candida albicans

Test suspension

Test - suspension			(N and No)	
N	Vc1	Vc2	x mean	2.82E+07
10 -6	24	32	log N	7.45
10 -7	3	3	No (N/10)	2.82E+06
			log No	6.45
			6,17 < = logNo < = 6,70	
				Yes

Validation and controls

Validation suspension (Nvo)			Experimental conditions (A)			Neutralizer control (B)			Method validation (C) Product conc.: undiluted (80%)		
Vc 1	64	x mean	Vc 1	71	x mean	Vc 1	67	x mean	Vc 1	61	x mean
Vc 2	67	65.5	Vc 2	62	66.5	Vc 2	62	64.5	Vc 2	64	62.5
30<x mean of Nvo < 160?			x mean of A is > 0,5*x mean of Nvo?			x mean of B is > 0,5*x mean of Nvo ?			x mean of C is > 0,5*x mean of Nvo?		
Yes			Yes			Yes			Yes		

Test Results

Product concentration (%)	Contact time	Dilution step	Vc 1	Vc 2	Average of Vc1 and Vc2	Na= average x10	log Na	log No	log Reduction (No-Na)	Criteria	Result
		10 0	0	0							
undiluted (80%)	15 seconds	10 -1	0	0	< 14	< 140	< 2.15	6.45	> 4.30	≥ 4	PASS TEST
50%	15 seconds	10 0	0	0	< 14	< 140	< 2.15	6.45	> 4.30	≥ 4	PASS TEST
		10 -1	0	0							
1%	15 seconds	10 -1	> 330	> 330	> 33000	> 330000	> 5.52	6.45	< 0.93	≥ 4	FAILS TEST
		10 -2	> 330	> 330							

Test Results for Aspergillus brasiliensis

Test suspension

Test - suspension			(N and No)	
N	Vc1	Vc2	x mean	1.82E+07
10 -6	19	17	log N	7.26
10 -7	2	2	No (N/10)	1.82E+06
			log No	6.26
			6,17 < = logNo < = 6,70	
				Yes

Validation and controls

Validation suspension (Nvo)			Experimental conditions (A)			Neutralizer control (B)			Method validation (C) Product conc.: undiluted (80%)		
Vc 1	34	x mean	Vc 1	35	x mean	Vc 1	40	x mean	Vc 1	37	x mean
Vc 2	39	36.5	Vc 2	41	38	Vc 2	36	38	Vc 2	43	40
30<x mean of Nvo < 160?			x mean of A is > 0,5*x mean of Nvo?			x mean of B is > 0,5*x mean of Nvo ?			x mean of C is > 0,5*x mean of Nvo?		
Yes			Yes			Yes			Yes		

Test Results

Product concentration (%)	Contact time	Dilution step	Vc 1	Vc 2	Average of Vc1 and Vc2	Na= average x10	log Na	log No	log Reduction (No-Na)	Criteria	Result
		10 0	> 165	> 165							
undiluted (80%)	15 seconds	10 -1	> 165	> 165	> 1650	> 16500	> 4.22	6.26	< 2.04	≥ 4	FAILS TEST
50%	15 seconds	10 0	> 165	> 165	> 1650	> 16500	> 4.22	6.26	< 2.04	≥ 4	FAILS TEST
		10 -1	> 165	> 165							

QACS LAB
QUALITY ASSURANCE & CONTROL SYSTEMS

A: 1 Antigonis str, 14451, Athens, Greece
T: (+30) 210 2934745 | E: info@qacs.gr
F: (+30) 210 2934606 | www.qacslab.com

PRODUCT NAME	:	Active chlorine released from hypochlorous acid by electrochemical by activation
ACTIVE SUBSTANCES	:	Active chlorine released from hypochlorous acid 0,05%
APPEARANCE OF THE PRODUCT	:	Liquid
STORAGE CONDITIONS	:	Room temperature, darkness
TEST CONDITIONS	:	Test conducted at 20°C □ 1 °C
LOT	:	02 – bacteria 1
METHOD	:	EN 1650:2019
RECEIPT DATE	:	17/02/2021
STUDY PERIOD	:	03/03/2021-05/03/2021
LAB ID	:	2021-1701/21 23 00161

1%	15 seconds	10^{0}	> 165	> 165	> 1650	> 16500	> 4.22	6.26	< 2.04	≥ 4	FAILS TEST
		10^{-1}	> 165	> 165							

CONCLUSION TEST PRODUCT IDENTIFICATION

METHODOLOGY ABSTRACT

A test suspension of fungi is tested against a product test solution at three concentrations with the presence of interfering substance. The mixture is maintained at 20 °C for 15 sec. At the end of this contact time, an aliquot is taken, and the fungicidal and/or yeasticidal activity in this portion is immediately neutralized or suppressed. The numbers of surviving fungi in each sample are determined and the reduction is calculated.

RESULT

The product under test: "Active chlorine released from hypochlorous acid by electrochemical by activation" demonstrated yeasticidal activity (≥ 4 log reduction) according to EN 1650:2019, at 20□1 °C, under clean conditions when tested at concentration:

Undiluted (80%) for 15 seconds contact time using as test organisms the reference strain: *Candida albicans*.

A: 1 Antigonis str, 14451, Athens, Greece
T: (+30) 210 2934745 | E: info@qacs.gr
F: (+30) 210 2934606 | www.qacslab.com

For the QACS Ltd Laboratory,

Signature date: 31/03/2021
Lagiopoulos Giorgos

PRODUCT NAME	:	Active chlorine released from hypochlorous acid by electrochemical by activation
ACTIVE SUBSTANCES	:	Active chlorine released from hypochlorous acid 0,05%
APPEARANCE OF THE PRODUCT	:	Liquid
STORAGE CONDITIONS	:	Room temperature, darkness
TEST CONDITIONS	:	Test conducted at 20°C □ 1 °C
LOT	:	02 – bacteria 1
METHOD	:	EN 1650:2019
RECEIPT DATE	:	17/02/2021
STUDY PERIOD	:	03/03/2021-05/03/2021
LAB ID	:	2021-1701/21 23 00161

Agronomist-Food Technologist M.Sc.
Study Manager

STUDY SUMMARY / ABSTRACT SUSPENSION TEST ACCORDING TO EN 1650:2019

Chemical disinfectants and antiseptics – evaluation of fungicidal and/or yeasticidal activity of chemical disinfectants and antiseptics used in food, industrial, domestic and institutional areas – Test method and requirements (Phase 2 step 1)

TEST PRODUCT IDENTIFICATION

TEST MICROORGANISMS

Candida albicans	ATCC 10231
Aspergillus brasiliensis	ATCC 16404

RESULT

The product under test: “Active chlorine released from hypochlorous acid by electrochemical by activation” demonstrated yeasticidal activity (≥ 4 log reduction) according to EN 1650:2019, at 20□1 °C, under clean conditions when tested at concentration:

Undiluted (80%) for 15 seconds contact time using as test organisms the reference strain: *Candida albicans*.

A: 1 Antigonis str, 14451, Athens, Greece
T: (+30) 210 2934745 | E: info@qacs.gr
F: (+30) 210 2934606 | www.qacslab.com

Results refer to the sample as received and analyzed on the period specified above.
The test report shall not be reproduced except in full, without written approval of the laboratory.
The samples will be stored by the laboratory during 1 month from the end test date.
The study report and raw data will be stored by the laboratory for 5 years.

End of Test Report