HAMILTON

AB 079

REPORT OF ANALYSIS No. 136987/21/JSHR/Z2

Replaces Report of Analysis No. 136987/21/JSHR of 2021-04-14

Client	Sample description *(according to declaration of Client)*
ECOCHIM-GRUP SRL OR. OTACI, STR. VOITOVICI 21 2059 CHIȘINĂU	**DEZINFECTANT UNIVERSAL "BIO-DEZ"** **Sample quantity: 2 pcs x 1 L** **Production date: 26.01.2021** **Expiration date: 26.01.2024** **Sampling date: 22.02.2021** **Sample temperature: 15°C** **Reception hour: 15:00** **Responsible for sampling: Crestinov Alexandr**
Sample received: **2021-03-18**	**Sample condition with no objections**
Analysis completed (the date of performance of the laboratory activity): **2021-04-14**	**Order of 2021-03-09**
Report dated: **2021-07-27**	The samples were delivered by Client

Test	Method	Unit	Result
* Chemical disinfectants and antiseptics - Hygienic handrub - Test method and requirements (phase 2, step 2)[1)]	PN-EN 1500:2013-07		The preparation has bactericidal effect against transient microorganisms used in the hygienic procedure of hand disinfection - a single rubbing of 3ml of the preparation for 60 seconds.

[1)] The results of the analysis in attachment No 1 to the report of analysis.

Identification of the change: test result

THE END OF THE REPORT

Digitally signed by Poiata Cristian
Date: 2023.03.31 13:08:45 EEST
Reason: MoldSign Signature
Location: Moldova

Authorized by:
Approved by: Hanna Wachowska, Laboratory Director *(Approved with electronic signature)*

Laboratory: Tychy 43-100, Goździków 1

* Test method accredited; # Test performed by external provider

J.S. HAMILTON POLAND Sp. z o.o.
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ENCLOSURE No. 1 TO REPORT OF ANALYSIS NO. 136987/21/JSHR/Z2

A) IDENTYFICATION OF THE SAMPLE:	
Name of the product	**DEZINFECTANT UNIVERSAL "BIO-DEZ"** Sample quantity: 2 pcs x 1 L Production date: 26.01.2021 Expiration date: 26.01.2024 Sampling date: 22.02.2021 Sample temperature: 15°C Reception hour: 15:00 Responsible for sampling: Crestinov Alexandr
The active substance	Ethyl alcohol 72-76% CAS 64-17-5 CE 200-578-6 Benzalkonium chloride 0,024-0,029% CAS 68424-85-1 CE 270-325-2 Methylthioninium chloride 0,00024% CAS 61-73-4 and 200-515-2
B) TEST METHOD :	
Method	**EN 1500:2013 Chemical disinfectants and antiseptics - Hygienic handrub - Test method and requirements (*phase 2, step 2*)**
Neutralizer	Polysorbate 80 30 g/l, saponine 30g/l, histidine 1g/l, cysteine 1g/l
C) EXPERIMENTAL CONDITIONS:	
Product test concentrations (%V/V)	100%
Test temperature	20°C
Contact time	3ml of the preparation for 60s
Incubation temperature	36±1 °C
Test-organism	*E. coli* K12 NCTC 10538

Date: 27.07.2021

Authorized by: Daria Depa, Senior Specialist Analyst, Cosmetics Microbiology Laboratory
Approved by: Hanna Wachowska, Laboratory Director *(Approved with qualified electronic signature)*

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ENCLOSURE No. 1 TO REPORT OF ANALYSIS NO. 136987/21/JSHR/Z2

Table 1. PROCEDURE FOR REFERENCE HYGIENIC HANDRUB

PRODUCT: Standard 2-propanol 60% (V/V)
TEST ORGANISM: *E. coli* K12 NCTC 10538
NUMBER IN CONTAMINATION FLUID: 2,4 x 10^8 cfu/ml

volunteer		number of cfu per plate from dilution 10x							
	Hand	prevalues			postvalues				Reduction
Nr	left/right	x10^{-4}	x10^{-5}	log x	x10^{0}	x10^{-1}	x 10^{-2}	log y	log z
	l	288	29		61	7	0		
1	r	247	22	6,42	33	3	0	1,65	4,77
	l	167	17		51	5	0		
2	r	291	28	5,81	36	4	0	1,63	4,18
	l	175	11		42	5	0		
3	r	275	25	6,33	29	2	0	1,54	4,79
	l	220	21		30	3	0		
4	r	192	19	6,31	68	6	0	1,65	4,66
	l	164	15		37	3	0		
5	r	301	33	6,35	52	5	0	1,64	4,71
	l	200	20		23	2	0		
6	r	198	18	6,30	37	4	0	1,46	4,83
	l	287	22		60	6	0		
7	r	288	29	6,45	42	5	0	1,70	4,75
	l	298	28		31	4	0		
8	r	213	21	6,40	58	5	0	1,63	4,77
	l	283	23		34	3	0		
9	r	311	33	5,96	51	5	0	1,62	4,34
	l	313	32		53	6	0		
10	r	251	25	6,45	36	4	0	1,65	4,80
	l	175	18		54	5	0		
11	r	295	22	6,35	47	3	0	1,69	4,66
	l	183	19		72	7	0		
12	r	171	17	5,74	36	4	0	1,71	4,03
	l	206	22		29	2	0		
13	r	317	33	6,41	49	5	0	1,57	4,84
	l	295	28		55	6	0		
14	r	279	25	6,45	64	7	0	1,78	4,68
	l	248	22		72	7	0		
15	r	256	26	6,40	66	6	0	1,84	4,56
	l	301	31		46	5	0		
16	r	261	26	6,45	27	3	0	1,55	4,90
	l	259	24		41	4	0		
17	r	271	28	6,42	22	1	0	1,47	4,96
	l	259	22		61	6	0		
18	r	288	23	6,43	33	3	0	1,65	4,78
	l	223	21		35	4	0		
19	r	205	20	6,33	45	5	0	1,60	4,72
	l	297	28		54	6	0		
20	r	257	24	5,90	28	3	0	1,59	4,31
$x_{śr}$				6,28				1,63	4,65
s				0,23				0,09	0,25

log x-logarithm of the average value of the initial left and right hand
log y-logarithm of the average value of the final left and right hand
log z-logarithm reduction
x śr- overall average of log x, log y, log z

Date: 27.07.2021

Authorized by: Daria Depa, Senior Specialist Analyst, Cosmetics Microbiology Laboratory
Approved by: Hanna Wachowska, Laboratory Director *(Approved with qualified electronic signature)*

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ENCLOSURE No. 1 TO REPORT OF ANALYSIS NO. 136987/21/JSHR/Z2

Table 2. HYGIENIC HANDRUB PROCEDURE WITH THE PRODUCT

PRODUCT P 136987/21/JSHR
TEST ORGANISM: *E. coli* K12 NCTC 10538
NUMBER IN CONTAMINATION FLUID: 2,4 x 10^8 cfu/ml

volunteer		number of cfu per plate from dilution 10x							
	Hand	prevalues			postvalues				Reduction
Nr	left/right	x10^{-4}	x10^{-5}	log x	x10^{0}	x10^{-1}	x 10^{-2}	log y	log z
1	l	132	14		103	11	1		
	r	224	21	6,24	92	9	0	1,98	4,26
2	l	>330	125		89	7	0		
	r	304	31	6,27	78	4	0	1,68	4,59
3	l	144	15		97	9	0		
	r	132	11	6,14	78	5	0	1,93	4,21
4	l	328	34		87	8	0		
	r	>330	85	6,20	99	9	0	1,89	4,32
5	l	164	11		116	11	2		
	r	132	12	6,16	99	8	0	2,03	4,13
6	l	>330	121		61	3	0		
	r	320	32	6,27	83	9	0	1,67	4,60
7	l	328	33		61	4	0		
	r	288	29	6,49	71	7	0	1,81	4,68
8	l	>330	58		91	9	0		
	r	>330	22	5,51	72	6	0	1,82	3,69
9	l	336	36		79	8	0		
	r	>330	21	5,90	106	12	2	1,96	3,94
10	l	296	28		74	7	0		
	r	>330	41	6,02	85	9	0	1,90	4,12
11	l	228	21		93	8	0		
	r	104	11	6,19	80	5	0	1,93	4,26
12	l	>330	48		107	11	1		
	r	200	20	5,97	94	9	0	1,98	4,00
13	l	248	25		112	14	2		
	r	212	22	6,36	113	11	1	2,06	4,31
14	l	>330	48		89	8	0		
	r	255	22	6,02	91	9	0	1,95	4,07
15	l	278	28		99	7	0		
	r	169	17	6,34	67	6	0	1,77	4,57
16	l	178	11		104	11	1		
	r	255	25	6,32	69	7	0	1,93	4,39
17	l	274	28		79	8	0		
	r	231	24	6,40	107	12	2	1,97	4,44
18	l	225	22		92	9	0		
	r	183	19	6,31	66	7	0	1,89	4,42
19	l	199	17		53	5	0		
	r	252	23	6,35	89	8	0	1,83	4,51
20	l	266	22		97	9	0		
	r	231	21	6,39	68	7	0	1,91	4,48
$x_{śr}$				6,19				1,89	4,30
s				0,22				0,10	0,25

log x-logarithm of the average value of the initial left and right hand
log y-logarithm of the average value of the final left and right hand
log z-logarithm reduction
x śr- overall average of log x, log y, log z

Date: 27.07.2021

Authorized by: Daria Depa, Senior Specialist Analyst, Cosmetics Microbiology Laboratory
Approved by: Hanna Wachowska, Laboratory Director *(Approved with qualified electronic signature)*

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ENCLOSURE No. 1 TO REPORT OF ANALYSIS NO. 136987/21/JSHR/Z2

Table 3. LIST OF COMPUTED IG VALUES AND IG REDUCTIONS

volunteer		R 2-propanol 60% (V/V)			P		
Nr		log x	log y	log z	log x	log y	log z
1	R-P	6,42	1,65	4,77	6,24	1,99	4,25
2	R-P	5,81	1,63	4,18	6,27	1,91	4,36
3	R-P	6,33	1,54	4,79	6,14	1,93	4,21
4	R-P	6,31	1,65	4,66	6,20	1,96	4,24
5	R-P	6,35	1,64	4,71	6,16	2,03	4,13
6	P-R	6,30	1,46	4,83	6,27	1,84	4,43
7	P-R	6,45	1,70	4,75	6,49	1,81	4,68
8	P-R	6,40	1,63	4,77	5,51	1,90	3,61
9	P-R	5,96	1,62	4,34	5,90	1,96	3,94
10	P-R	6,45	1,65	4,80	6,02	1,90	4,12
11	R-P	6,35	1,69	4,66	6,19	1,93	4,26
12	R-P	5,74	1,71	4,03	5,97	2,00	3,97
13	R-P	6,41	1,57	4,84	6,36	2,06	4,31
14	R-P	6,45	1,78	4,68	6,02	1,95	4,07
15	R-P	6,40	1,84	4,56	6,34	1,90	4,43
16	P-R	6,45	1,55	4,90	6,32	1,93	4,39
17	P-R	6,42	1,47	4,96	6,40	1,97	4,44
18	P-R	6,43	1,65	4,78	6,31	1,89	4,42
19	P-R	6,33	1,60	4,72	6,35	1,83	4,51
20	P-R	5,90	1,59	4,31	6,39	1,91	4,48
X_{20}		**6,28**	1,63	4,65	**6,19**	1,93	4,26
X10(R-P)		6,26	1,67	**4,59**	6,19	1,97	**4,22**
X10 (P-R)		6,31	1,59	**4,72**	6,20	1,90	**4,30**

Criteria:
Rs (R-P) = 4,59-4,22=0,37
Rs (P-R)= 4,72-4,30=0,42
Abs= 0,37-0,42=-0,05<2
logx(R)= 6,28>5
logx(P)= 6,19>5
logz (P), logz (R) >3

Validation conditions of neutralizer and methods have been satisfied

Date: 27.07.2021

Authorized by: Daria Depa, Senior Specialist Analyst, Cosmetics Microbiology Laboratory
Approved by: Hanna Wachowska, Laboratory Director *(Approved with qualified electronic signature)*

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Table 4. COMPUTATION OF INDIVIDUAL DIFFERENCES OF lg R-P

volunteer	log RF		difference	difference	
	R	P	R-P	high to low	Range +/-
1	4,77	4,25	0,52	1,16	1
2	4,18	4,36	-0,18	0,68	2
3	4,79	4,21	0,59	0,61	3
4	4,66	4,24	0,42	0,59	4
5	4,71	4,13	0,58	0,58	5
6	4,83	4,43	0,40	0,53	6
7	4,75	4,68	0,07	0,52	7
8	4,77	3,61	1,16	0,52	8
9	4,34	3,94	0,40	0,51	9
10	4,80	4,12	0,68	0,42	10
11	4,66	4,26	0,40	0,40	11
12	4,03	3,97	0,06	0,40	12
13	4,84	4,31	0,53	0,40	13
14	4,68	4,07	0,61	0,36	14
15	4,56	4,43	0,13	0,21	15
16	4,90	4,39	0,51	0,13	16
17	4,96	4,44	0,52	0,07	17
18	4,78	4,42	0,36	0,06	18
19	4,72	4,51	0,21	-0,17	-19
20	4,31	4,48	-0,17	-0,18	-20
sum of ranks (+): 171					
sum of ranks (-): 39					

Table 5. SORTING OF INDIVIDUAL DIFFERENCES AND COMPUTATION FOR HODGES-LEHMANN 97,5% UPPER CONFIDENCE LIMITS FOR THE DIFFERENCE IN lg BETWEEN R-P

		1,16	0,68	0,61	0,59	0,58	0,53	0,52	0,52	0,51
1	1,16	1,16								
2	0,68	0,92	0,68							
3	0,61	0,89	0,65	0,61						
4	0,59	0,87	0,63	0,60	0,59					
5	0,58	0,87	0,63	0,59	0,58	0,58				
6	0,53	0,85	0,61	0,57	0,56	0,55	0,53			
7	0,52	0,84	0,60	0,57	0,56	**0,55**	0,53	-0,52		
8	0,52	0,84	0,60	0,56	0,55	0,55	0,53	-0,52	-0,52	
9	0,51	0,83	0,59	0,56	0,55	0,54	0,52	-0,52	-0,51	-0,51
10	0,42	0,79	0,55	0,52	0,50	0,50	0,48	-0,47	-0,47	-0,21
11	0,40	0,78	0,54	0,51	0,49	0,49	0,47	-0,46	-0,46	-0,20
12	0,40	0,78	0,54	0,51	0,49	0,49	0,47	-0,46	-0,46	
13	0,40	0,78	0,54	0,50	0,49	0,49	0,47	-0,46		
14	0,36	0,76	0,52	0,49	0,47	0,47	0,45			
15	0,21	0,69	0,45	0,41	0,40	0,39				
16	0,13	0,65	0,41	0,37	0,36					
17	0,07	0,62	0,38	0,34						
18	0,06	0,61	0,37							
19	-0,17	0,50								
20	-0,18									

Date: 27.07.2021

Authorized by: Daria Depa, Senior Specialist Analyst, Cosmetics Microbiology Laboratory
Approved by: Hanna Wachowska, Laboratory Director *(Approved with qualified electronic signature)*

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ENCLOSURE No. 1 TO REPORT OF ANALYSIS NO. 136987/21/JSHR/Z2

Table 6. WILCOXON'S TMATCHED PAIRS SIGNED-RANKS TEST:
CRITICAL VALUES LESS WITH RANG SUM (+) OR (-) AT DIFFERENT LEVELS OF SIGNIFICANCE

n	one-sided level of significance		
	0,05	**0,025**	0,01
18	47	**40**	32
19	53	**46**	27
20	60	**52**	43
21	68	**59**	49
22	75	**66**	56

For the designated level of significance 0,025 for n=20 the value read from the table 6 is 52.
Hence c = 52+1 =53.
For the distribution of 53 Table 5 assigns a value of 0,55 which is less than the agreed inferiority margin of 0,6.
Therefore, the hypothesis of inferiority of PP compared to the reference RP is rejected.
The test preparation (PP) is non-inferior to RP.

Date: 27.07.2021

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HAMILTON ROMANIA

RAPORT DE INCERCARE NR. 79165/22/ROBCH/Z1

Inlocuieste Raportul de Incercare Nr. 79165/22/ROBCH din 07.03.2022

Client	Numărul eșantionului:
ECOCHIM-GRUP SRL OR. OTACI, STR. VOITOVICI 21 2059 CHIȘINĂU	Sample number Descriere obiect de incercat (conform cu declaratia Clientului) Dezinfectant Universal "Bio-Dez" Lot/Batch: - Production date: - Expiration date: 18.02.2025 Sampling date: 18.02.2022 Sampling quantity: 1x 500ml Sample temperature: 20°C Reception hour: 12:30 Responsible for sampling: Crestinov Alexandr Sample condition with no objections
Data primirii obiectului de incercat: 07.03.2022	
Data finalizarii incercarii: 08.09.2022	Comanda din 07.03.2022
Data eliberarii raportului: 08.09.2022	Probele au fost prelevate si livrate de catre Client.

Parametrii de testare	Metoda de testare	Unitate de masura	Rezultat
# * Chemical disinfectants and antiseptics. Surgical hand disinfection. Test method and requirements (phase 2, step 2)	EN 12791:2016+A1:2017	-	Test method performed by the subcontractor; the results are taken in full from the test report No S68/2022-1, issued by the subcontractor. The results of the analysis are listed starting with enclosure No1 to report of analysis.

Elaborat de: Mariana Ilinca, Sef Laborator Microbiologie
Autorizat de: Mariana Ilinca, Sef Laborator Microbiologie
Aprobat de catre: Alina-Roxana Mihai, General Manager (Aprobat cu semnatura electronica)

Laborator: Bucuresti 041914, sos. Berceni nr.8

* Metoda de testare acreditata # Test efectuat de catre subcontractor
ø Incercari neacreditate

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Autorizatie A.N.S.V.S.A. nr. 115 din 18.06.2018

ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 79165/22/ROBCH

A) IDENTYFICATION OF THE SAMPLE	
Name of the product/Details about the product	**Dezinfectant Universal "Bio-Dez"** Date of manufacture: 18.02.2022 Manufacturer: Ecochim-Grup SRL, Petricani St 21/3, Chișinău 2059, Moldavsko Incoming date: 7.3.2022 Storing conditions: room temperature, dark area. Subject of testing: Surgical handrub - immediate effect Active ingredients: CAS: 68424-85- Alkyldimethylbenzylamoniumchloride 0.029% CAS: 64-17-5 Ethyl alcohol 73.0%
B) TEST METHOD	
Performed in accredited subcontracted partner laboratory: Scope of Accreditation No. 1273 Determination of surgical hand disinfection (EN 12791:2016+A1:2017) of the product **Dezinfectant Universal "Bio-Dez"**	Testing of disinfecting efficiency of chemical disinfecting and antiseptic agents on carriers SOP-M-19-00 (EN 12791:2016+A1:2017)
C) Description: Testing the efficacy of chemical disinfectants and antiseptics	
Sampling date:	18.2.2022
Sample delivered:	7.3.2022
Testing date:	16.8. - 31.8.2022
D) EXPERIMENTAL CONDITIONS	
Effect:	immediate effect
Period of analysis:	16.08.2022 - 31.08.2022
Test temperature:	20 °C ± 1 °C
Test method:	dilution neutralization method
Appearance of the product:	Blue liquid
The test concentration:	100%
The volume of the product:	2 x 7 ml
The application time:	2 x 45 s
Procedure:	handrub
The soap:	soft soap from linseed oil
Reference item:	CAS 71-23-8 1-propanol p.a., 60% (V/V)
The volume of the reference propan- -1-ol used per person: 2 x 3 ml, according to reference surgical hand disinfection procedure, the total application volume is 6 ml	
The application time:	2 x 1.5 min, according to reference surgical hand disinfection procedure, the total application time is 3 min
Neutralization medium:	Dey-Engley Neutralizing Broth M 1062
Surgical hand disinfection procedure with product:	handrub procedure, immediate effect
Requirements:	The mean reduction for immediate effect of a product shall at least be not inferior to that achieved by a specified reference product (60% volume concentration of propan-1-ol). To demonstrate additionally a "sustained effect", the mean reduction for the 3 h effect of a product shall be superior to that achieved by the reference product
Test procedure:	1. Determination of the presence of microorganisms in the product 2. Determination of the prevalue – number of cfu sampled immediately before treatment from the hand 3. Determination of the postvalue – number of cfu sampled after

	treatment from the hand 4. Expression and interpretation of results - reduction factor – ratio of prevalue and postvalue, generally expressed by decimal logarithms
The standard:	EN 12791:2016+A1:2017 Chemical disinfectants and antiseptics – Surgical hand disinfection – Test method and requirements (phase 2/step 2) November 2017

The number of CFU in the tested product: 0 CFU/ml

Testing the efficacy of chemical disinfectant **L21229/22/JSHR** *on Pseudomonas aeruginosa* ATCC 15442

Test suspensions

N	V1	V2	lgN	lgN_0
10^{-6}	192	170		
10^{-7}	27	15	8,26	7,26
		$\Phi = 1,84x10^{8}$	*8,17* ≤ lgN ≤ *8,7*	*7,17* ≤ lgN_0 ≤ *7,7*

Verification of methodology

Validation of suspension (N_{V0})		Method valid.(C), conditions: 80 %, 90 s, distilled water, 20°C	
V_{c1}	31	V_{c1}	29
V_{c2}	60	V_{c2}	55
30 < 45,5 < 160		42 > 0,5 Nvo	

Testing the efficacy of chemical disinfectant **L21229/22/JSHR** *on Staphylococcus aureus* ATCC 6538

Test suspensions

N	V1	V2	lgN	lgN_0
10^{-6}	304	252		
10^{-7}	20	33	8,44	7,44
		$\Phi = 2,77x10^{8}$	*8,17* < lgN < *8,7*	*7,17* < lgN_0 < *7,7*

Verification of methodology

Validation of suspension (N_{V0})		Method valid.(C), conditions: 80 %, 90 s, distilled water, 20°C	
V_{c1}	69	V_{c1}	72
V_{c2}	53	V_{c2}	42
30 < 61 < 160		57 > 0,5 Nvo	

Testing the efficacy of chemical disinfectant **L21229/22/JSHR** *on Enterococcus hirae* ATCC 10541

Test suspensions

N	V1	V2	lgN	lgN_0
10^{-6}	166	192		
10^{-7}	18	21	8,26	7,26
		$\Phi = 1,8x10^{8}$	*8,17* < lgN < *8,7*	*7,17* < lgN_0 < *7,7*

Verification of methodology

Validation of suspension (N_{V0})		Method valid.(C), conditions: 80 %, 90 s, distilled water, 20°C	
V_{c1}	52	V_{c1}	46
V_{c2}	40	V_{c2}	42
30 < 46 < 160		44 > 0,5 Nvo	

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ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 79165/22/ROBCH

Testing the efficacy of chemical disinfectant **L21229/22/JSHR** *on Escherichia coli K 12* NCTC 10538

Test suspensions

N	V1	V2	lgN	lgN$_0$
10^{-6}	>330	>330		
10^{-7}	49	33	8,61	7,61
		$\Phi = 4,1x10^{8}$	8,17 < lgN < 8,7	7,17 < lgN$_0$ < 7,7

Verification of methodology

Validation of suspension (Nv0)		Method valid.(C), conditions: 80 %, 90 s, distilled water, 20°C	
V_{c1}	113	V_{c1}	103
V_{c2}	103	V_{c2}	110
30 < 108 < 160		106,5 > 0,5 Nvo	

Testing the efficacy of chemical disinfectant **L21229/22/JSHR** *on Candida albicans* ATCC 10231

Test suspensions

N	V1	V2	lgN	lgN$_0$
10^{-5}	155	144		
10^{-6}	17	15	7,18	6,18
		$\Phi = 1,5x10^{7}$	7,17 ≤ lgN ≤ 7,7	7,17 ≤ lgN$_0$ ≤ 7,7

Verification of methodology

Validation of suspension (Nv0)		Method valid.(C), conditions: 80 %, 90 s, distilled water, 20°C	
V_{c1}	41	V_{c1}	21
V_{c2}	34	V_{c2}	28
30 < 37,5 < 160		24,5 > 0,5 Nvo	

Note: Vc = value is the number of cfu per ml, Φ = average Vc1 a Vc2 (1. + 2. duplicate Vc values), N = the number of cfu/ml of the bacterial test suspension, Nv0 = the number of cfu/ml of the bacterial test suspension at the beginning of the contact time = 0, C = the number of surviving bacteria per ml in control tests

Acceptance criteria for test results:

Only if the results of the test procedure fulfil the following requirements, they shall be accepted for further evaluation, otherwise the test shall be repeated:

a) A complete set of results from at least 23, but maximum 28 volunteers shall be available. All complete sets of results shall be used for further evaluation.

b) The overall means of the lg prevalues for RP and PP shall be both at least 3,50.

c) The absolute difference of mean differences between lg reductions of RP and PP of group RP → PP and group PP → RP shall be less than 2,00

d) All quotients of weighted mean counts between 5 and 15.

*Test method accredited # Test performed by subcontractor. Ø Non accredited methods.

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Date: 08.09.2022
Authorized by: Mariana Ilinca, Manager of Microbiological laboratory
Aprobat de: Mihai Alina-Roxana, General manager (Approved with qualified electronic signature)

ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 79165/22/ROBCH

Conclusion:

The acceptance criteria for the test results were met.
From table in EN 12791:2016+A1:2017 of critical values for Wilcoxon's matched-pairs signed-ranks test the entry for n = 24 and a one sided 0,025 level of significance, the critical value of 81 is found. **Hence c = 81 +1 = 82**. The pairwise differences are sorted in descending order. **The 82nd value is 0,59.** Hence the Hodges-Lehmann upper one sided 97,5% confidence limit for the difference in lg Rs between RP and PP is 0,59, which is less than the agreed inferiority margin of 0,75. Therefore the hypothesis of inferiority of PP is rejected and it can be concluded that the test preparation PP is non-inferior to RP.

The tested product:	**Dezinfectant Universal "Bio-Dez"**
Batch number:	Not specified
Standard:	EN 12791:2016+A1:2017
Test method:	dilution neutralization method
Effect:	immediate effect

Conditions:

Application time:	2 x 45 s
Volume of the product:	2 x 7 ml
Concentration:	100%

The tested product is suitable to be used as surgical hand disinfection.

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Annex

EN 12791:2016+A1:2017,
immediate effect

Bio-Dez
sample S68/2022

Volunteer	Chronological Sequence	Reference hand disinfection procedure RP					Reference handrub procedure with product PP					Difference RP - PP
		N prevalues	N postvalues	lg prevalues	lg postvalues	lg R	N prevalues	N postvalues	lg prevalues	lg postvalues	lg R	
1	RP	7,30E+04	7,90E+02	4,86	2,90	1,96	6,40E+04	7,70E+02	4,81	2,89	1,92	0,04
2	RP	7,90E+04	6,60E+03	4,90	3,82	1,08	9,00E+04	1,00E+04	4,95	4,00	0,95	0,13
3	RP	7,10E+04	1,88E+03	4,85	3,27	1,58	1,06E+05	1,20E+03	5,03	3,08	1,95	-0,37
4	RP	7,90E+04	8,50E+02	4,90	2,93	1,97	5,80E+04	1,00E+02	4,76	2,00	2,76	-0,79
5	RP	9,60E+04	9,60E+03	4,98	3,98	1,00	8,90E+04	1,12E+04	4,95	4,05	0,90	0,10
6	RP	8,50E+04	2,44E+03	4,93	3,39	1,54	1,70E+04	6,70E+03	4,23	3,83	0,40	1,14
7	RP	7,70E+04	7,10E+03	4,89	3,85	1,04	1,02E+04	2,21E+03	4,01	3,34	0,67	0,37
8	RP	6,80E+04	3,80E+02	4,83	2,58	2,25	1,29E+05	9,30E+03	5,11	3,97	1,14	1,11
9	RP	5,60E+04	4,90E+03	4,75	3,69	1,06	8,30E+04	5,10E+03	4,92	3,71	1,21	-0,15
10	RP	7,10E+04	9,90E+03	4,85	4,00	0,85	7,60E+04	9,40E+03	4,88	3,97	0,91	-0,06
11	RP	1,02E+05	1,01E+04	5,01	4,00	1,01	6,90E+04	9,30E+03	4,84	3,97	0,87	0,14
12	RP	9,40E+04	1,07E+04	4,97	4,03	0,94	1,11E+05	1,24E+04	5,05	4,09	0,96	-0,02
13	PP	7,90E+04	2,91E+03	4,90	3,46	1,44	9,80E+04	7,80E+03	4,99	3,89	1,10	0,34
14	PP	9,10E+04	6,10E+03	4,96	3,79	1,17	7,80E+04	8,00E+03	4,89	3,90	0,99	0,18
15	PP	7,50E+04	3,08E+03	4,88	3,49	1,39	1,12E+04	1,56E+03	4,05	3,19	0,86	0,53
16	PP	5,20E+04	9,30E+02	4,72	2,97	1,75	1,10E+05	2,01E+02	5,04	2,30	2,74	-0,99
17	PP	8,50E+04	6,50E+03	4,93	3,81	1,12	1,02E+05	1,23E+04	5,01	4,09	0,92	0,20
18	PP	1,06E+04	2,04E+02	4,03	2,31	1,72	6,50E+03	7,50E+02	3,81	2,88	0,93	0,79
19	PP	8,00E+04	1,10E+03	4,90	3,04	1,86	1,68E+04	4,80E+02	4,23	2,68	1,55	0,31
20	PP	1,10E+05	6,00E+03	5,04	3,78	1,26	9,90E+04	8,60E+03	5,00	3,93	1,07	0,19
21	PP	6,30E+04	2,76E+02	4,80	2,44	2,36	8,70E+04	2,86E+03	4,94	3,46	1,48	0,88
22	PP	5,50E+04	4,40E+02	4,74	2,64	2,10	5,30E+04	7,10E+03	4,72	3,85	0,87	1,23
23	PP	1,18E+05	9,70E+02	5,07	2,99	2,08	8,20E+04	6,50E+03	4,91	3,81	1,10	0,98
24	PP	1,04E+05	7,60E+02	5,02	2,88	2,14	1,16E+05	1,35E+04	5,06	4,13	0,93	1,21
Ø	Overall	7,81E+04	3,94E+03	4,86	3,34	1,52	7,34E+04	6,14E+03	4,76	3,54	1,22	
s		2,23E+04	3,62E+03	0,20	0,55	0,48	3,68E+04	4,43E+03	0,38	0,61	0,59	
n				24	24	24			24	24	24	
Ø	RP → PP			4,89	3,54	1,35			4,79	3,57	1,22	0,13
s				0,07	0,51	0,48			0,33	0,64	0,66	
n				12	12	12			12	12	12	
Ø	PP → RP			4,83	3,13	1,70			4,72	3,51	1,21	0,49
s				0,28	0,53	0,42			0,44	0,61	0,53	
n				12	12	12			12	12	12	

Sorting of individual differences and computation for Hodges-Lehman 97,5% upper confidence limits													
	Sorted differences	**Mean pairwise differences (di+dii)/2**											
1	1,23	1,23											
2	1,21	1,22	1,21										
3	1,14	1,19	1,18	1,14									
4	1,11	1,17	1,16	1,13	1,11								
5	0,98	1,11	1,10	1,06	1,05	0,98							
6	0,88	1,06	1,05	1,01	1,00	0,93	0,88						
7	0,79	1,01	1,00	0,97	0,95	0,89	0,84	0,79					
8	0,53	0,88	0,87	0,84	0,82	0,76	0,71	0,66	0,53				
9	0,37	0,80	0,79	0,76	0,74	0,68	0,63	0,58	0,45	0,37			
10	0,34	0,79	0,78	0,74	0,73	0,66	0,61	0,57	0,44	0,36	0,34		
11	0,31	0,77	0,76	0,73	0,71	0,65	0,60	0,55	0,42	0,34	0,33	0,31	
12	0,20	0,72	0,71	0,67	0,66	0,59	0,54	0,50	0,37	0,29	0,27	0,26	0,20
13	0,19	0,71	0,70	0,67	0,65	0,59	0,54	0,49	0,36	0,28	0,27	0,25	0,20
14	0,18	0,71	0,70	0,66	0,65	0,58	0,53	0,49	0,36	0,28	0,26	0,25	
15	0,14	0,69	0,68	0,64	0,63	0,56	0,51	0,47	0,34	0,26	0,24		
16	0,13	0,68	0,67	0,64	0,62	0,56	0,51	0,46	0,33	0,25			
17	0,10	0,67	0,66	0,62	0,61	0,54	0,49	0,45	0,32				
18	0,04	0,64	0,63	0,59	0,58	0,51	0,46	0,42					
19	-0,02	0,61	0,60	0,56	0,55	0,48	0,43						
20	-0,06	0,59	0,58	0,54	0,53	0,46							
21	-0,15	0,54	0,53	0,50	0,48								
22	-0,37	0,43	0,42	0,39									
23	-0,79	0,22	0,21										
24	-0,99	0,12											

log R = decimal log reduction; RP→PP sequence: first RP, second PP; PP→RP sequence: first PP, second RP; Ø = mean; s = standard deviation; n = number of values (volunteer)

Difference of mean Rs (RP→PP): 1,35 - 1,22 = 0,13; Difference of mean Rs (PP→RP): 1,70 - 1,21 = 0,49 ; Absolute difference of differences: |0,13 - 0,49| = 0,36

The median is between the 12th and 13th value: [0,20 + 0,19]/2 = 0,195

The mean pairwise differences that do not exceed the median (here: 0,195) are computed. From table in EN 12791:2016+A1:2017 of critical values for Wilcoxon's matched-pairs signed-ranks test the entry for n = 24 ans a one-sided P = 0,025 level of significance, the critical value of 81 is found. **Hence c = 81 + 1 =82.** The pairwise differences are sorted in descending order. **The 82nd value is 0,59.**

Hence the Hodges-Lehmann upper one sided 97,5% confidence limit for the difference in lg Rs between RP and PP is 0**,59,** which is less than the agreed inferiority margin of 0,75.

Therefore the hypothesis of inferiority of PP is rejected and it can be concluded that the test preparation PP is non-inferior to RP.

Propan-1-ol batch No.: K52972497115, expiry date 31.12.25 60%, 2 x 3 ml, 2 x 1,5 min, immediate effect, hand disinfection procedure

Volunteer			Number of CFU per plate from dilution 10^x						
No	Sequence	Hand (left or right)	Prevalues			Immediate postvalues			
			-1	-2	-3	0	-1	-2	
1	RP → PP	l	>330	>330	73	>330	79	<14	Period of analysis: 16.8.2022 - 31.8.2022
2	RP → PP	l	>330	>330	79	>330	>330	66	16.8.-17.8.2022, 30.8.-31.8.2022
3	RP → PP	l	>330	>330	71	>330	186	21	Prepared by: Mgr. Alena Holíková
4	RP → PP	l	>330	>330	79	>330	85	<14	
5	RP → PP	l	>330	>330	96	>330	>330	96	
6	RP → PP	l	>330	>330	85	>330	241	27	
7	RP → PP	r	>330	>330	77	>330	>330	71	
8	RP → PP	r	>330	>330	68	>330	38	<14	
9	RP → PP	r	>330	>330	56	>330	>330	49	
10	RP → PP	r	>330	>330	71	>330	>330	99	
11	RP → PP	r	>330	>330	102	>330	>330	101	
12	RP → PP	r	>330	>330	94	>330	>330	107	
13	PP → RP	l	>330	>330	79	>330	292	28	
14	PP → RP	l	>330	>330	91	>330	>330	61	
15	PP → RP	l	>330	>330	75	>330	306	33	
16	PP → RP	l	>330	>330	52	>330	93	<14	
17	PP → RP	l	>330	>330	85	>330	>330	65	
18	PP → RP	l	>330	106	<14	201	23	<14	
19	PP → RP	r	>330	>330	80	>330	110	<14	
20	PP → RP	r	>330	>330	110	>330	>330	60	
21	PP → RP	r	>330	>330	63	269	35	<14	
22	PP → RP	r	>330	>330	55	>330	44	<14	
23	PP → RP	r	>330	>330	118	>330	97	<14	
24	PP → RP	r	>330	>330	104	>330	76	<14	

Product Dezinfectant Universal "Bio-Dez" 100%, 2 x 7 ml, 2 x 45 s, immediate effect, handrub

Volunteer			Number of CFU per plate from dilution 10^x						
No	Sequence	Hand (left or right)	Prevalues			Immediate postvalues			
			-1	-2	-3	0	-1	-2	
1	RP → PP	r	>330	>330	64	>330	77	<14	Period of analysis: 16.8.2022 - 31.8.2022
2	RP → PP	r	>330	>330	90	>330	>330	101	16.8.-17.8.2022, 30.8.-31.8.2022
3	RP → PP	r	>330	>330	106	>330	120	<14	Prepared by: Mgr. Alena Holíková
4	RP → PP	r	>330	>330	58	99	<14	<14	
5	RP → PP	r	>330	>330	89	>330	>330	112	
6	RP → PP	r	>330	168	19	>330	>330	67	
7	RP → PP	l	>330	102	<14	>330	216	27	
8	RP → PP	l	>330	>330	129	>330	>330	93	
9	RP → PP	l	>330	>330	83	>330	>330	51	
10	RP → PP	l	>330	>330	76	>330	>330	94	
11	RP → PP	l	>330	>330	69	>330	>330	93	
12	RP → PP	l	>330	>330	111	>330	>330	124	
13	PP → RP	r	>330	>330	98	>330	>330	78	
14	PP → RP	r	>330	>330	78	>330	>330	80	
15	PP → RP	r	>330	112	<14	>330	156	16	
16	PP → RP	r	>330	>330	110	198	23	<14	
17	PP → RP	r	>330	>330	102	>330	>330	123	
18	PP → RP	r	>330	65	<14	>330	75	<14	
19	PP → RP	l	>330	164	21	>330	48	<14	
20	PP → RP	l	>330	>330	99	>330	>330	86	
21	PP → RP	l	>330	>330	87	>330	284	31	
22	PP → RP	l	>330	>330	53	>330	>330	71	
23	PP → RP	l	>330	>330	82	>330	>330	65	
24	PP → RP	l	>330	>330	116	>330	>330	135	

HAMILTON ROMANIA

REPORT OF ANALYSIS No. 80248/21/ROBCH

Client	Sample number:
ECOCHIM-GRUP SRL OR. OTACI, STR. VOITOVICI 21 2059 CHIȘINĂU	**80248/21/ROBCH** Sample description *(according to declaration of Client)* **DEZINFECTANT UNIVERSAL "BIO-DEZ"** **Lot: -** **Data fabricatie: 01.10.2021** **Data expirarii: 01.10.2024** **Data prelevarii: -** **Cantitate prelevata: 2 x 500 ml** **Responsabil prelevare: CRESTINOV ALEXANDR** **Ora receptiei probei: 15:30** **Temperatura receptie proba: 15°C** **Sample condition with no objections**
Sample received: **11.10.2021**	
Tests performed: **21.10.2021**	
Tests completed: **13.12.2021**	**Order of 11.10.2021**
Report dated: **13.12.2021**	Sampling and delivery were carried out by client.

Test	Method	Unit	Result
# * Fungicidal activity in medical area. Chemical disinfectants and antiseptics. Quantitative suspension test for the evaluation of fungicidal or yeasticidal activity in the medical area. Test method and requirements (phase 2, step 1).	EN 13624:2014	-	Test method performed by the subcontractor; the results are taken in full from the test report No D/21/ B0644 , issued by the subcontractor. The results of the analysis are listed starting with enclosure No1 to report of analysis.

Elaborated by: Mariana Ilinca, Manager of Microbiological Laboratory
Authorized by: Mariana Ilinca, Manager of Microbiological Laboratory
Approved by: Alina-Roxana Mihai, General Manager *(Approved with qualified electronic signature)*

Laboratory: Bucuresti 041914, sos. Berceni nr.8

* Test method accredited # Test performed by external provider
ø Non accredited methods

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ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 80248/21/ROBCH

A) IDENTYFICATION OF THE SAMPLE	
Name of the product/Details about the product	**DEZINFECTANT UNIVERSAL "BIO-DEZ"** Expiration date: 01.10.2024. Manufacturer (supplier): Ecochim-Grup SRL. Storing conditions: Dry, without sun, 5-25 Celsius degree. Conditions of use: Hygienic handrub, surface disinfection, medical instruments disinfection, surgical handrub
Active compound/s and its concentration/s	Ethyl alcohol 72-76%, CAS 64-17-5 and CE 200-578-6. Benzalkonium chloride 0.024-0.029%, CAS 68424-85-1 and CE 270-325-2, Methylthionibium chloride 0.00024%, CAS 61-73-4 and 200-515-2
Concentrations requested for the assay	Pure (80%).
B) TEST METHOD	
Performed in accredited contracted partner laboratory, Scope of Accreditation Nr. 648/LE1286 Report Registration No. D/21/B0644 Quantitative evaluation assay of yeasticidal activity under dirty conditions, in the medical area (phase 2, step 1), with product Desinfectant Universal"Bio-Dez", (UNE-EN 13624: 2014 Standard).	UNE-EN 13624:2014 Chemical disinfectants and antiseptics. Quantitative suspension test for the evaluation of the fungicidal or yeasticidal activity in the medical area. Test method and requirements (phase 2, step 1). AENOR.
Testing method	Procedure DESIN-1058-b // EN 13624:2014
C) INFORMATION ABOUT SAMPLE RECEPTION	
Date of reception of order with test conditions	21.10.2021
Date of reception of the sample	25.10.2021
Aspect of the received product	Blue liquid in plastic package
D) METHOD OF ASSAY AND ITS VALIDATION (UNE-EN 13624: 2014 Standard)	
Method used	Dilution-neutralization
Neutralizer	Tryptone 5 g/L, yeast extract 2.5 g/L, dextrose 10 g/L, sodium thioglycolate 1 g/L, sodium thiosulfate 1 g/L, sodium bisulphite 2.5 g/L, soy lecithin 7 g/L, polysorbate-80 5 g/L, glycine 1 g/L, l-histidine 1 g/L and Saponin 30 g/L.
E) EXPERIMENTAL CONDITIONS	
Assay period	2021/11/08 to 2021/11/14.
Solvent of the product used in the assay	Sterile distilled water.
Product concentrations for the assay	Pure (80%), 50%, 0.1%
Aspect of the dilutions of the product	Pure (80%) and 50% blue liquid; 0.1% transparent.
Contact time	60 seconds
Assay temperature	+20°C ± 1°C
Interfering substance	Bovine serum albumin 3 g/L and erythrocytes 3 mL/L.
Stability of the mixture (interfering substance and product diluted in sterile distilled water)	Stable
Temperature of incubation	+30°C± 1°C
Identification of the strain used	*Candida albicans* CECT-1394 (ATCC 10231)

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Date: 08.12.2021
Authorized by: Mariana Ilinca, Manager of Microbiological laboratory
Aprobat de: Mihai Alina-Roxana, General manager (Approved with qualified electronic signature)

ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 80248/21/ROBCH

Results of the assay

- Assay of validation ..... See tables 1 and 2.
- Evaluation of yeasticidal activity..... See table 3.
- Number of replicates per assay organism ..... 1.

Special remarks

- All controls and validation were between the basic limits.
- At least one concentration of the sample showed a log reduction lower than 4 log.
- At least one concentration of the sample showed a log reduction higher than 4 log.
- The highest concentration that can be assayed in the test (80%) is due to the mixtures to perform the assay.
- No precipitate formed during the test procedure (the test mixtures were homogeneous).

Conclusion

The product **Desinfectant Universal"Bio-Dez"**, batch not indicated, when is pure (80%), shows **yeasticidal activity** after 60 seconds at 20ºC ± 1ºC, under dirty conditions (bovine serum albumin 3 g/L and erythrocytes 3 mL/L), for the reference strain *Candida albicans* (CECT 1394 = ATCC 10231), when tested as required by the **UNE-EN 13624: 2014 Standard,**.

Note: The results obtained correspond to the sample received in the laboratory.

Quality Assurance Review:

The assay development and the results obtained have been supervised by the Director of the study.

The Quality Assurance Director has inspected the development of the assay, proving that has been realized following the proper procedure and using the adequate media, materials, and reagents, following the Good Laboratory Practices (GLPs) as well and the final report contains the primary data obtained.

Reference

- **UNE-EN 13624 : 2014**. Chemical disinfectants and antiseptics. Quantitative suspension test for the evaluation of the fungicidal or yeasticidal activity in the medical area. Test method and requirements (phase 2, step 1). AENOR.

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ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 80248/21/ROBCH

Results of the assay with *Candida albicans* (CECT 1394 = ATCC 10231).

Seeding: Pour plates. No. of plates: 1 /mL.

Table 1.-Validation and controls.

Suspension of validation (Nv_0)			Control of experimental conditions (A)			Control of neutralizer (B)			Validation of the method (C) Sample concentration: Pure (80%)		
Vc_1	86	X= 90	Vc_1	72	X= 74	Vc_1	75	X= 73	Vc_1	66	X=
Vc_2	94		Vc_2	76		Vc_2	71		Vc_2	61	63.5
30 ≤ x of Nv_0 ≤ 160? Yes			x of A es ≥ 0,5 X de Nv_0? Yes			x of B es ≥ 0,5 X de Nv_0, or 0.0005 Nv_B? Yes			x of C es ≥ 0.5 X of Nv_0? Yes		
Suspension of validation (Nv_B)			Vc_1: 79 Vc_2: 77			X = 78 30 ≤ x de Nv_B/1000 ≤ 160? Yes					

Table 2. -Suspension of the assay.

	N	Vc_1	Vc_2	$Xwm = 3.35 \times 10^7$
Suspension of assay (N and N_0)	10^{-5}	>330	>330	lg N = 7.53 $N_0 = N/10$
	10^{-6}	32	35	lg N_0 = 6.53 6.17 ≤ lg N_0 ≤ 6.70? Yes

Table 3.-Results of the activity assays with the sample.

Concentrations of the sample (%)	Dilutions steps	Vc_1	Vc_2	Lg Na = lg (X x 10 or Xmw x 10)	LgR (lg N_0 = 6.53)	Time of contact (seconds)
Pure (80%)	Na^0	<14	<14	<2.15	>4.38	60
	Na^{-1}	<14	<14			
50%	Na^0	<14	<14	<2.15	>4.38	60
	Na^{-1}	<14	<14			
0.1%	Na^0	>330	>330	>4.52	<2.01	60
	Na^{-1}	>330	>330			

Explanations:

Vc = number per mL (one or two plates); Xwm = ponderated mean of X.

X = mean of Vc_1 and Vc_2 (duplicate of 1 + 2); R (reduction): (lg R = lg N_0 – lg Na).

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Date: 08.12.2021
Authorized by: Mariana Ilinca, Manager of Microbiological laboratory
Aprobat de: Mihai Alina-Roxana, General manager (Approved with qualified electronic signature)

HAMILTON ROMANIA

REPORT OF ANALYSIS No. 80249/21/ROBCH

Client **ECOCHIM-GRUP SRL** OR. OTACI, STR. VOITOVICI 21 2059 CHIȘINĂU	Sample number: **80249/21/ROBCH** Sample description *(according to declaration of Client)* **DEZINFECTANT UNIVERSAL "BIO-DEZ"** **Lot: -** **Data fabricatie: 01.10.2021** **Data expirarii: 01.10.2024** **Data prelevarii: -** **Cantitate prelevata: 1 x 500 ml** **Responsabil prelevare: CRESTINOV ALEXANDR** **Ora receptiei probei: 15:30** **Temperatura receptie proba: 15°C** **Sample condition with no objections**
Sample received: **11.10.2021** Tests performed: **21.10.2021** Tests completed: **13.12.2021** Report dated: **13.12.2021**	**Order of 11.10.2021** Sampling and delivery were carried out by client.

Test	Method	Unit	Result
# * Quantitative suspension test for the evaluation of bactericidal activity in medical area	EN 13727:2012+A2:2015	-	Test method performed by the subcontractor; the results are taken in full from the test report No D/21/ B0645, issued by the subcontractor. The results of the analysis are listed starting with enclosure No1 to report of analysis.

Elaborated by: Mariana Ilinca, Manager of Microbiological Laboratory
Authorized by: Mariana Ilinca, Manager of Microbiological Laboratory
Approved by: Alina-Roxana Mihai, General Manager *(Approved with qualified electronic signature)*

Laboratory: Bucuresti 041914, sos. Berceni nr.8

* Test method accredited # Test performed by external provider
ø Non accredited methods

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ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 80249/21/ROBCH

A) IDENTYFICATION OF THE SAMPLE	
Name of the product/Details about the product	**DEZINFECTANT UNIVERSAL "BIO-DEZ"** Expiration date: 01.10.2024. Manufacturer (supplier): Ecochim-Grup SRL. Storing conditions: Dry, without sun, 5-25 Celsius degree. Conditions of use: Hygienic handrub, surface disinfection, medical instruments disinfection, surgical handrub
Active compound/s and its concentration/s	Ethyl alcohol 72-76%, CAS 64-17-5 and CE 200-578-6. Benzalkonium chloride 0.024- 0.029%, CAS 68424-85-1 and CE 270-325-2, Methylthionibium chloride 0.00024%, CAS 61-73-4 and 200-515-2
Concentrations requested for the assay	Pure (80%).
B) TEST METHOD	
Performed in accredited contracted partner laboratory, Scope of Accreditation Nr. 648/LE1286 Report Registration No. D/21/B0645 Quantitative evaluation assay of the bactericidal activity under dirty conditions, in the medical area (phase 2, step 1) with product Desinfectant Universal"Bio-Dez", (UNE-EN 13727: 2012 + A2: 2015 Standard).	UNE-EN 13727: 2012 + A2: 2015. Chemical disinfectants and antiseptics. Quantitative suspension test for the evaluation of bactericidal activity of chemical disinfectants for instruments used in Medicine. Test method and requirements (phase 2, step 1). AENOR.
Testing method	DESIN-1031-b //EN 13727: 2012 + A2: 2015
C) INFORMATION ABOUT SAMPLE RECEPTION	
Date of reception of order with test conditions	21.10.2021
Date of reception of the sample	25.10.2021
Aspect of the received product	Blue liquid in plastic package
D) METHOD OF ASSAY AND ITS VALIDATION (UNE-EN 13727: 2012+A2: 2015 Standard.)	
Method used	Dilution-neutralization
Neutralizer	Tryptone 5 g/L, yeast extract 2.5 g/L, dextrose 10 g/L, sodium thioglycolate 1 g/L, sodium thiosulfate 1 g/L, sodium bisulphite 2.5 g/L, soya lecithin 7 g/L, polysorbate-80 5 g/L, glycine 1 g/L, l-histidine 1 g/L and saponin 30 g/L.
E) EXPERIMENTAL CONDITIONS	
Assay period	2021/11/10 to 2021/11/14.
Solvent of the product used in the assay	Sterile distilled water.
Product concentrations for the assay	Pure (80%), 50%, 0.1%
Aspect of the dilutions of the product	Pure (80%) and 50% blue liquid; 0.1% transparent.
Contact time	60 seconds
Assay temperature	+20°C ± 1°C
Interfering substance	Bovine serum albumin 3 g/L and erythrocytes 3 mL/L.
Stability of the mixture (interfering substance and product diluted in sterile distilled water)	Stable
Temperature of incubation	+36°C± 1°C
Identification of the strain used	− *Pseudomonas aeruginosa* (CECT 116 = ATCC 15442). − *Staphylococcus aureus* (CECT 239 = ATCC 6538). − *Enterococcus hirae* (CECT 4081 = ATCC 10541). − *Escherichia coli K12* (CECT 433 = NCTC 10538).

Date: 08.12.2021
Authorized by: Mariana Ilinca, Manager of Microbiological laboratory
Aprobat de: Mihai Alina-Roxana, General manager (Approved with qualified electronic signature)

ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 80249/21/ROBCH

Results of the assay

- Assay of validation ..... See tables 1, 2, 4, 5, 7, 8, 10 and 11.
- Evaluation of bactericidal activity..... See tables 3, 6, 9 and 12.
- Number of replicates per assay organism .. 1.

Special remarks

- All controls and validation were between the basic limits.
- At least one concentration of the sample showed a log reduction lower than 5 log.
- At least one concentration of the sample showed a log reduction higher than 5 log.
- The highest concentration that can be assayed in the test (80%) is due to the mixtures to perform the assay.
- No precipitate formed during the test procedure (the test mixtures were homogeneous).

Conclusion

The product **Desinfectant Universal"Bio-Dez"**, batch not indicated, when is pure (80%), **shows bactericidal activity** after 60 seconds at 20ºC ± 1ºC, under dirty conditions (bovine serum albumin 3 g/L and erythrocytes 3 mL/L), for the reference strains *Pseudomonas aeruginosa* (CECT 116 = ATCC 15442), *Staphylococcus aureus* (CECT 239 = ATCC 6538), *Enterococcus hirae* (CECT 4081 = ATCC 10541) and *Escherichia coli* K12 (CECT 433 = NCTC 10538), when tested according to **UNE-EN 13727: 2012 + A2: 2015 Standard**.

Note: The results obtained correspond to the sample received in the laboratory.

Quality Assurance Review:

The assay development and the results obtained have been supervised by the Director of the study.

The Quality Assurance Director has inspected the development of the assay, proving that has been realized following the proper procedure and using the adequate media, materials, and reagents, following the Good Laboratory Practices (GLPs) as well and the final report contains the primary data obtained.

Reference

- **UNE-EN 13727: 2012 + A2: 2015.** Chemical disinfectants and antiseptics. Quantitative suspension test for the evaluation of bactericidal activity of chemical disinfectants for instruments used in Medicine. Test method and requirements (phase 2, step 1). AENOR.

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ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 80249/21/ROBCH

Results of the assay (Bactericidal suspension) with *Pseudomonas aeruginosa* (CECT 116 = ATCC 15442).

Seeding: Pour plate; No. of plates: 1/mL.

Table 1.-Validation and controls

Suspension of validation (Nv_0)			Control of experimental conditions (A)			Control of neutralizer (B)			Validation of the method (C) Sample concentration: Pure (80%)		
Vc_1	61	X=	Vc_1	53	X= 54	Vc_1	46	X= 48	Vc_1	42	X=
Vc_2	56	58.5	Vc_2	55		Vc_2	50		Vc_2	37	39.5
30 ≤ x of Nv_0 ≤ 160? Yes			x of A es ≥ 0,5 X de Nv_0? Yes			x of B es ≥ 0,5 X de Nv_0, or 0.0005 Nv_B? Yes			x of C es ≥ 0.5 X of Nv_0? Yes		
Suspension of validation (Nv_B)			Vc_1: 57 Vc_2: 63			X = 60 30 ≤ x de Nv_B/1000 ≤ 160? Yes					

Table 2.-Suspension of the assay

	N	Vc_1	Vc_2	
Suspension of assay (N and N_0)	10^{-6}	217	234	Xwm = 2.25 x 10^8, lg N = 8.35 N_0 = N/10; lg N_0 = 7.35
	10^{-7}	21	22	7.17 ≤ lg N_0 ≤ 7.70? Yes

Table 3.-Results of the activity assays with the sample

Concentrations of the sample (%)	Dilutions steps	Vc_1	Vc_2	Lg Na = lg (X x 10 o Xwm x 10)	Lg R (Lg N_0=7.35)	Time of contact (seconds)
Pure (80%)	Na^0	<14	<14	<2.15	>5.20	60
	Na^{-1}	<14	<14			
50%	Na^0	<14	<14	<2.15	>5.20	60
	Na^{-1}	<14	<14			
0.1%	Na^0	>330	>330	>4.52	<2.83	60
	Na^{-1}	>330	>330			

Explanations:

Vc = number per mL (one or two plates); Xwm = weighted mean of X.

X = mean of Vc_1 and Vc_2 (duplicate of 1 + 2);

Logarithmic reduction R: ($\lg R = \lg N_0 - \lg Na$).

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ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 80249/21/ROBCH

Results of the assay (Bactericidal suspension) with *Staphylococcus aureus* (CECT 239 = ATCC 6538).

Seeding: Pour plate; No. of plates: 1/mL.

Table 4.-Validation and controls

Suspension of validation (Nv_0)			Control of experimental conditions (A)			Control of neutralizer (B)			Validation of the method (C) Sample concentration: Pure (80%)		
Vc_1	77	X= 74	Vc_1	68	X=	Vc_1	75	X=	Vc_1	70	X= 66
Vc_2	71		Vc_2	69	68.5	Vc_2	82	78.5	Vc_2	62	
30 ≤ x of Nv_0 ≤ 160? Yes			x of A es ≥ 0,5 X de Nv_0? Yes			x of B es ≥ 0,5 X de Nv_0, or 0.0005 Nv_B? Yes			x of C es ≥ 0.5 X of Nv_0? Yes		
Suspension of validation (Nv_B)			Vc_1: 79 Vc_2: 86			X = 82.5 30 ≤ x de Nv_B/1000 ≤ 160? Yes					

Table 5.-Suspension of the assay

	N	Vc_1	Vc_2	
Suspension of assay (N and N_0)	10^{-6}	>330	>330	Xwm = 3.40 x 10^8, lg N = 8.53 N_0 = N/10; lg N_0 = 7.53 7.17 ≤ lg N_0 ≤ 7.70? Yes
	10^{-7}	33	35	

Table 6.-Results of the activity assays with the sample

Concentrations of the sample (%)	Dilutions steps	Vc_1	Vc_2	Lg Na = lg (X x 10 o Xwm x 10)	Lg R (Lg N_0=7.53)	Time of contact (seconds)
Pure (80%)	Na^0	<14	<14	<2.15	>5.38	60
	Na^{-1}	<14	<14			
50%	Na^0	<14	<14	<2.15	>5.38	60
	Na^{-1}	<14	<14			
0.1%	Na^0	>330	>330	>4.52	<3.01	60
	Na^{-1}	>330	>330			

Explanations:
Vc = number per mL (one or two plates); Xwm = weighted mean of X.
X = mean of Vc_1 and Vc_2 (duplicate of 1 + 2);
Logarithmic reduction R: (lg$R$ = lg N_0 – lg Na).

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ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 80249/21/ROBCH

Results of the assay (Bactericidal suspension) with *Enterococcus hirae* (CECT 4081 = ATCC 10541).

Seeding: Pour plate; No. of plates: 1/mL.

Table 7.-Validation and controls

Suspension of validation (Nv_0)			Control of experimental conditions (A)			Control of neutralizer (B)			Validation of the method (C) Sample concentration: Pure (80%)		
Vc_1	41	X= 43	Vc_1	44	X=	Vc_1	37	X=	Vc_1	35	X= 36
Vc_2	45		Vc_2	43	41.5	Vc_2	40	38.5	Vc_2	37	
30 ≤ x of Nv_0 ≤ 160? Yes			x of A es ≥ 0,5 X de Nv_0? Yes			x of B es ≥ 0,5 X de Nv_0, or 0.0005 Nv_B? Yes			x of C es ≥ 0.5 X of Nv_0? Yes		
Suspension of validation (Nv_B)			Vc_1: 39 Vc_2: 42			X = 40.5 30 ≤ x de Nv_B/1000 ≤ 160? Yes					

Table 8.-Suspension of the assay

	N	Vc_1	Vc_2	
Suspension of assay (N and N_0)	10^{-6}	167	154	Xwm = 1.61 x 10^8, lg N = 8.20 N_0 = N/10; lg N_0 = 7.20 7.17 ≤ lg N_0 ≤ 7.70? Yes
	10^{-7}	17	16	

Table 9.-Results of the activity assays with the sample

Concentrations of the sample (%)	Dilutions steps	Vc_1	Vc_2	Lg Na = lg (X x 10 o Xwm x 10)	Lg R (Lg N_0=7.20)	Time of contact (seconds)
Pure (80%)	Na^0	<14	<14	<2.15	>5.05	60
	Na^{-1}	<14	<14			
50%	Na^0	15	14	2.16	5.04	60
	Na^{-1}	<14	<14			
0.1%	Na^0	>330	>330	>4.52	<2.68	60
	Na^{-1}	>330	>330			

Explanations:
Vc = number per mL (one or two plates); Xwm = weighted mean of X.
X = mean of Vc_1 and Vc_2 (duplicate of 1 + 2);
Logarithmic reduction R: (lg$R$ = lg N_0 – lg Na).

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ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 80249/21/ROBCH

Results of the assay (Bactericidal suspension) with *Escherichia coli K12* (CECT 433 = NCTC 10538).

Seeding: Pour plate; No. of plates: 1/mL.

Table 10.-Validation and controls

Suspension of validation (Nv_0)			Control of experimental conditions (A)			Control of neutralizer (B)			Validation of the method (C) Sample concentration: Pure (80%)		
Vc_1	58	X= 60	Vc_1	44	X= 45	Vc_1	51	X= 50	Vc_1	43	X= 41
Vc_2	62		Vc_2	46		Vc_2	49		Vc_2	39	
30 ≤ x of Nv_0 ≤ 160? Yes			x of A es ≥ 0,5 X de Nv_0? Yes			x of B es ≥ 0,5 X de Nv_0, or 0.0005 Nv_B? Yes			x of C es ≥ 0.5 X of Nv_0? Yes		
Suspension of validation (Nv_B)			Vc_1: 54 Vc_2: 56			X = 55 30 ≤ x de Nv_B/1000 ≤ 160? Yes					

Table 11.-Suspension of the assay

	N	Vc_1	Vc_2	
Suspension of assay (N and N_0)	10^{-6}	241	259	Xwm = 2.49 x 10^8, lg N = 8.40 N_0 = N/10; lg N_0 = 7.40 7.17 ≤ lg N_0 ≤ 7.70? Yes
	10^{-7}	22	25	

Table 12.-Results of the activity assays with the sample

Concentrations of the sample (%)	Dilutions steps	Vc_1	Vc_2	Lg Na = lg (X x 10 o Xwm x 10)	Lg R (Lg N_0=7.40)	Time of contact (seconds)
Pure (80%)	Na^0	<14	<14	<2.15	>5.25	60
	Na^{-1}	<14	<14			
50%	Na^0	<14	<14	<2.15	>5.25	60
	Na^{-1}	<14	<14			
0.1%	Na^0	>330	>330	>4.52	<2.88	60
	Na^{-1}	>330	>330			

Explanations:
Vc = number per mL (one or two plates); Xwm = weighted mean of X.
X = mean of Vc_1 and Vc_2 (duplicate of 1 + 2);
Logarithmic reduction R: (lg$R$ = lg N_0 – lg Na).

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Date: 08.12.2021
Authorized by: Mariana Ilinca, Manager of Microbiological laboratory
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REPORT OF ANALYSIS No. 17898/21/ROBCH

Client	Sample number:
ECOCHIM-GRUP SRL OR. OTACI, STR. VOITOVICI 21 2059 CHIȘINĂU	**17898/21/ROBCH** Sample description *(according to declaration of Client)* **DEZINFECTANT UNIVERSAL "BIO-DEZ"** **Sample quantity: 1 pcs x 1 L** **Production date: 26.01.2021** **Expiration date: 26.01.2024** **Sampling date: 22.02.2021** **Sample temperature: 15°C** **Reception hour: 15:00** **Responsible for sampling: Crestinov Alexandr**
Sample received: **15.03.2021**	**Sample condition with no objections**
Tests performed: **21.04.2021**	
Tests completed: **16.06.2021**	**Order of 15.03.2021**
Report dated: **16.06.2021**	Sampling and delivery were carried out by client.

Test	Method	Unit	Result
# * Chemical disinfectants and antiseptics. Quantitative suspension test for the evaluation of mycobactericidal activity of chemical disinfectants in the medical area including instrument disinfectants. Test methods and requirements (phase 2, step 1)	EN 14348: 2005	-	Test method performed by the subcontractor; the results are taken in full from the test report No D/21/ B0152, issued by the subcontractor. The results of the analysis are listed starting with enclosure No1 to report of analysis.

Elaborated by: Mariana Ilinca, Manager of Microbiological Laboratory
Authorized by: Mariana Ilinca, Manager of Microbiological Laboratory
Approved by: Alina-Roxana Mihai, General Manager *(Approved with qualified electronic signature)*

Laboratory: Bucuresti 041914, sos. Berceni nr.8

* Test method accredited # Test performed by external provider
ø Non accredited methods

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ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 17898/21/ROBCH

A) IDENTYFICATION OF THE SAMPLE	
Name of the product/Details about the product	**DEZINFECTANT UNIVERSAL "BIO-DEZ"** Expiration date: 26.01.2024 Manufacturer (supplier): ECOCHIM-GRUP Storing conditions: Dry, without sun, 5-25 Celsius degree. Conditions of use: Handrub
Active(S) substance (S) and its concentration(s)	Ethyl alcohol 72-76%, CAS 64-17-5 and CE 200-578-6; Benzalkonium chloride 0.024-0.029%, CAS 68424-85-1 and CE 270-325-2; Methylthioninium chloride 0.00024%, CAS 61-73-4 and 200
Concentrations requested for the assay	3%/ on May 5 the client requested to perform the test at 80% concentration (Pure).
B) TEST METHOD	
Performed in accredited subcontracted partner laboratory: Scope of Accreditation Nr. 648/LE1286 Report D/21/B0152 Mycobactericidal and tuberculocidal activity of chemical disinfectants in the medical area including instrument disinfectants under clean conditions with the product **DEZINFECTANT UNIVERSAL "BIO-DEZ"** with deviations from the standard **(UNE-EN 14348: 2005 Standard)**	UNE-EN 14348: 2005. Chemical disinfectants and antiseptics . Quantitative suspension test for the evaluation of mycobactericidal activity of chemical disinfectants in the medical area including instrument disinfectants. Test methods and requirements (phase 2, step 1). AFNOR
C) METHOD OF ASSAY AND ITS VALIDATION (UNE-EN 14348: 2005 Standard)	
Testing method	DESIN-1052-b // EN 14348: 2005
Method used	Dilution-neutralization
Neutralizer	Tryptone 5 g/L, yeast extract 2.5 g/L, dextrose 10 g/L, sodium thioglycolate 1 g/L, sodium thiosulfate 1 g/L, sodium bisulphite 2.5 g/L, soya lecithin 7 g/L, polysorbate-80 5 g/L, glycine 1 g/L, l-histidine 1 g/L and saponim 30 g/L.
D) INFORMATION ABOUT SAMPLE RECEPTION	
Date of reception of the sample	24.03.2021
Date of reception of order with test conditions	14.04.2021: 3% concentration 05.05.2021: 80% concentration
Aspect of the received product	Blue liquid in plastic package.
E) EXPERIMENTAL CONDITIONS	
Assay period	2021/04/12 to 2021/05/24 (including prior preparation of the strains)
Solvent of the product used in the assay	Sterile hard water
Product concentrations for the assay	Pure (80%), 3% and 0.1%
Aspect of the dilutions of the product	Pure (80%) Blue liquid; 3% and 0.1% transparent
Contact time	60 seconds
Assay temperature	20°C ± 1°C
Interfering substance	Bovine albumin 0.3 g/L
Stability of the mixture (interfering substance and product diluted in sterile hard water)	stable
Temperature of incubation	36°C ± 1°C
Identification of the origin of viral stains and number of passes	*Mycobacterium avium* (ATCC 15769) *Mycobacterium terrae* (CECT 3028 = ATCC 15755)

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Date: 09.06.2021
Authorized by: Mariana Ilinca, Manager of Microbiological laboratory
Aprobat de: Mihai Alina-Roxana, General manager (Approved with qualified electronic signature)

ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 17898/21/ROBCH

Results of the assay

- Control and validation assays..... See tables 1, 2, 4 and 5
- Evaluation of mycobactericidal activity... See tables 3 and 6.
- Number of replicates for each assay microorganism..... 1.

Special remarks

- All controls and validation were between the basic limits.
- One concentration of the sample at least showed a log reduction less than 4 log.
- One concentration of the sample at least showed a log reduction higher than 4 log.
- The highest concentration that can be assayed in the test (80%) is due to the mixtures to perform the assay.
- When the client requested to perform the test at 80% concentration, the test had been started, using hard water.

Conclusion

The product **DEZINFECTANT UNIVERSAL „BIO-DEZ"**, batch not indicated, when tested pure (80%), **shows mycobactericidal activity** after 60 seconds at 20°C under clean conditions (bovine albumin 0.3 g/L), against *Mycobacterium avium* (ATCC 15769) and *Mycobacterium terrae* (CECT 3028 = ATCC 15755), when tested as required by **UNE-EN 14348: 2005 Standard** with deviations from the standard since the dilutions of the product, ready to use, have been prepared in sterile hard water instead of in sterile distilled water. The client informed us that the product was ready to use once the test have been started.

Note: The results obtained correspond to the sample received in the laboratory.

Quality Assurance Review:

The assay development and the results obtained have been supervised by the Director of the study.

The Quality Assurance Director has inspected the development of the assay, proving that has been realized following the proper procedure and using the adequate media, materials and reagents, following as well the Good Laboratory Practices (GLPs) and the final report contains the primary data obtained.

Reference:

- **UNE-EN 14348: 2005**. Chemical disinfectants and antiseptics. Quantitative suspension test for the evaluation of mycobactericidal activity of chemical disinfectants in the medical area including instrument disinfectants. Test methods and requirements (phase 2, step 1). AENOR.

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ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 17898/21/ROBCH

Table 1.-Assay with *Mycobacterium avium* (ATCC 15769): Validation and controls.

Suspension of validation (Nv_0)			Control of experimental conditions (A)			Neutralizer (B)			Validation of the method (C) with sample concentration: Pure (80%)		
Vc_1	135	X=	Vc_1	125	X= 122	Vc_1	121	X= 124	Vc_1	130	X=
Vc_2	124	129.5	Vc_2	119		Vc_2	127		Vc_2	119	124.5
30 ≤ x of Nv_0 ≤ 160? Yes			x of A is ≥ 0.5 x X of Nv_0? Yes			x of B is ≥ 0.5 x of Nv_0? Yes			x of C is ≥ 0.5 X of Nv_0? Yes		

Table 2.- Assay with *Mycobacterium avium* (ATCC 15769): Suspension of the assay.

Suspension of the assay (N y N_0)	N	Vc_1	Vc_2	$X_{wm} = 4.85 \times 10^9 = \lg = 9.69$ $N_0 = N/10 = \lg = 8.69$ $8.17 \leq N_0 \leq 8.70$? Yes
	10^{-7}	>660	>660	
	10^{-8}	51	46	

Table 3.- Assay with *Mycobacterium avium* (ATCC 15769).

Concentrations of the sample (%)	Dilutions	Vc_1	Vc_2	Lg Na = lg (X x 10 o Xwm x 10)	LgR (lgN_0= 8.69)	Time of contact (seconds)
Pure (80%)	10^0	<14	<14	<2.15	>6.54	60
	10^{-1}	<14	<14			
	10^{-2}	<14	<14			
	10^{-3}	<14	<14			
3%	10^0	>660	>660	>6.82	<1.87	60
	10^{-1}	>660	>660			
	10^{-2}	>660	>660			
	10^{-3}	>660	>660			
0.1%	10^0	>660	>660	>6.82	<1.87	60
	10^{-1}	>660	>660			
	10^{-2}	>660	>660			
	10^{-3}	>660	>660			

Observations:

N 10^{-7}: >330 + >330; >330 + >330;
10^{-8}: 28 + 23; 27 + 19;

Nv₀: 74 + 61; 59 + 65;
A: 68 + 57; 60 + 59;
B: 63 + 58; 71 + 57;
C: 72 + 58; 55 + 64;

Na Pure (80%) 10^0: 0 + 0; 0 + 0;
3% 10^{-3}: >330 + >330; >330 + >330;
0.1% 10^{-3}: >330 + >330; >330 + >330;

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Table 4.-Assay with *Mycobacterium terrae* (CECT 3028 = ATCC 15755): Validation and controls.

Suspension of validation (Nv_0)			Control of experimental conditions (A)			Neutralizer (B)			Validation of the method (C) with sample concentration: Pure (80%)		
Vc_1	53	X= 55	Vc_1	54	X= 52	Vc_1	50	X= 49	Vc_1	49	X= 47
Vc_2	57		Vc_2	50		Vc_2	48		Vc_2	45	
30 ≤ x of Nv_0 ≤ 160? Yes			x of A is ≥ 0.5 x X of Nv_0? Yes			x of B is ≥ 0.5 x of Nv_0? Yes			x of C is ≥ 0.5 X of Nv_0? Yes		

Table 5.- Assay with *Mycobacterium terrae* (CECT 3028 = ATCC-15755): Suspension of the assay.

Suspension of the assay (N y N_0)	N	Vc_1	Vc_2	$X_{wm} = 2.19 \times 10^9 = \lg = 9.34$ $N_0 = N/10 = \lg = 8.34$ $8.17 \leq N_0 \leq 8.70$? Yes
	10^{-7}	227	211	
	10^{-8}	21	22	

Table 6.- Assay with *Mycobacterium terrae* (CECT 3028 = ATCC 15755).

Concentrations of the sample (%)	Dilutions	Vc_1	Vc_2	Lg Na = lg (X x 10 o Xwm x 10)	LgR (lgN_0= 8.34)	Time of contact (seconds)
Pure (80%)	10^0	<14	<14	<2.15	>6.19	60
	10^{-1}	<14	<14			
	10^{-2}	<14	<14			
	10^{-3}	<14	<14			
3%	10^0	>660	>660	>6.82	<1.52	60
	10^{-1}	>660	>660			
	10^{-2}	>660	>660			
	10^{-3}	>660	>660			
0.1%	10^0	>660	>660	>6.82	<1.52	60
	10^{-1}	>660	>660			
	10^{-2}	>660	>660			
	10^{-3}	>660	>660			

Observations:

N 10^{-7}: 103 + 124; 99 + 112;
10^{-8}: 13 + 8; 10 + 12;

Na Pure (80%) 10^0: 0 + 0; 0 + 0;
3% 10^{-3}: >330 + >330; >330 + >330;
0.1% 10^{-3}: >330 + >330; >330 + >330;

Nv_0: 29 + 24; 32 + 25;
A: 31 + 23; 26 + 24;
B: 19 + 31; 27 + 21;
C: 33 + 16; 21 + 24;

Explanations:
Vc: Counts per mL
Xwm: ponderated mean of X
X: Values of Vc_1 and Vc_2 (1. + 2. duplicates); *R*: reduction (LgR = lgN_0 – lgNa)
R: reduction (LgR = lgN_0 – lgNa)

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Date: 09.06.2021
Authorized by: Mariana Ilinca, Manager of Microbiological laboratory
Aprobat de: Mihai Alina-Roxana, General manager (Approved with qualified electronic signature)

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*Test method accredited # Test performed by subcontractor. Ø Non accredited methods.

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Aprobat de: Mihai Alina-Roxana, General manager (Approved with qualified electronic signature)

HAMILTON ROMANIA

RAPORT DE INCERCARE NR. 17899/21/ROBCH/Z1

Inlocuieste Raportul de Incercare Nr. 17899/21/ROBCH din 26.04.2021

Client **ECOCHIM-GRUP SRL** OR. OTACI, STR. VOITOVICI 21 2059 CHIȘINĂU	Numă rul esantionului: Sample number Descriere obiect de incercat (conform cu declaratia Clientului) DEZINFECTANT UNIVERSAL "BIO-DEZ" Sample quantity: 1 pcs x 1 L Production date: 26.01.2021 Expiration date: 26.01.2024 Sampling date: 22.02.2021 Sample temperature: 15°C Reception hour: 15:00 Responsible for sampling: Crestinov Alexandr Sample condition with no objections
Data primirii obiectului de incercat: 15.03.2021	
Data finalizarii incercarii: 26.04.2021	Comanda din 15.03.2021
Data eliberarii raportului: 14.09.2021	Probele au fost prelevate si livrate de catre Client.

Parametrii de testare	Metoda de testare	Unitate de masura	Rezultat
# * Chemical disinfectants and antiseptics. Quantitative suspension test for the evaluation of virucidal activity in the medical area	UNE-EN 14476:2014 + A2:2019	-	Test method performed by the subcontractor; the results are taken in full from the test report No D/21/ V0204, issued by the subcontractor. The results of the analysis are listed starting with enclosure No1 to report of analysis.

Elaborat de: Mariana Ilinca, Sef Laborator Microbiologie
Autorizat de: Mariana Ilinca, Sef Laborator Microbiologie
Aprobat de catre: Alina-Roxana Mihai, General Manager (Aprobat cu semnatura electronica)

Laborator: Bucuresti 041914, sos. Berceni nr.8

* Metoda de testare acreditata # Test efectuat de catre subcontractor
ø Incercari neacreditate

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ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 17899/21/ROBCH/Z1

A) IDENTYFICATION OF THE SAMPLE	
Name of the product/Details about the product	**DEZINFECTANT UNIVERSAL "BIO-DEZ"** Sample quantity: 1 pcs x 1 L Production date: 26.01.2021 Expiration date: 26.01.2024 Manufacturer(supplier): ECOCHIM-GRUP Dry, without sun, 5-25^{0}C
Active(s) Substance(s) and its concentration(s)	Ethyl alcohol 72-76%, CAS 64-17-5 and CE 200-578-6; Benzalkonium chloride 0,024-0,029%, CAS 68424-85-1 and CE 270-325-2; Methylthioninium chloride 0,00024%, CAS 61-73-4 and 200.
Concentration ordered for the assay	3%
B) TEST METHOD	
Performed in accredited subcontracted partner laboratory: Scope of Accreditation Nr. 648/LE1286 Report D/21/V0204- Modified report Virucidal test with the sample **DEZINFECTANT UNIVERSAL "BIO-DEZ"** against Poliovirus type 1, Adenovirus type 5 and Murine Norovirus (NF EN 14476:2013+A2:2019 Standard)	NF EN 14476:2013+A2:2019 Standard. Virucidal quantitative suspension test for chemical disinfectants and antiseptics used in human medicine. Test method and requirements (Phase 2/Step1).AFNOR
Testing method	Procedure DESIN-1078 (NF EN 14476:2013+ A2:2019 Standard)
C) INFORMATION ABOUT SAMPLE RECEPTION	
Date of reception of the sample	22.03.2021
Date of reception of order with test conditions	22.03.2021
Aspect of the received product	Blue transparent liquid in a plastic container
D) EXPERIMENTAL CONDITIONS	
Assay period	From 22.03.2021 to 01.04.2021
Assay temperature	37°C ± 1°C
Titration method	TCID50 (Tissue Culture Infective Dose 50%)
Product concentrations for the assay	80%, 3% and 0.03%
Contact time	60 seconds
Contact temperature	20°C ± 1°C
Procedure to stop product cytotoxicity	Molecular sieving (< 4 columns)
Procedure to stop product activity	Cooling with ice
Solvent of the product used in the assay	Hard water
Aspect of the dilutions of the product	Transparent
Stability of the mixture (interfering substance and product diluted in sterile hard water/distilled water)	Stable
Interfering substance	Clean conditions in the presence of bovine serum albumin 0.3 g/L.
Identification of the origin of viral stains and number of passes	Poliovirus type 1 aliquot: 2021/01/07 passage 2; Adenovirus type 5 aliquot: 2021/01/14 passage 2; Murine Norovirus aliquot: 2021/02/11 passage 2;
Cell lines (name, origin, number of passes)	Vero ref: FTVE, working aliquot 12 passages 13 and 17; Raw 264.7, Public Health England, working aliquot 12, passages 12, 16 and 18;

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Note: Modified report due to the client requests to add the 80% concentration results in the conclusions section.

Validation of assay results

Poliovirus type 1 (ATCC VR-192)

Titre of the viral suspension for the virus control (at the requested test time):
- Clean conditions ..... $\log 10^{-7.24}$

Cytotoxicity level (80%) ..... $\log 10^{-0.50}$

Maximum level of virus inactivation detectable (difference between the titre of the viral suspension and the cytotoxicity level):
- Clean conditions ..... $\log 10^{-6.74}$

Adenovirus type 5 (ATCC VR-5)

Titre of the viral suspension for the virus control (at the requested test time):
- Clean conditions ..... $\log 10^{-6.33}$

Cytotoxicity level (80%) ..... $\log 10^{-0.50}$

Maximum level of virus inactivation detectable (difference between the titre of the viral suspension and the cytotoxicity level):
- Clean conditions ..... $\log 10^{-5.83}$

Murine Norovirus (strain S99 Berlin)

Titre of the viral suspension for the virus control (at the requested test time):
- Clean conditions ..... $\log 10^{-6.16}$

Cytotoxicity level (80%) ..... $\log 10^{-0.50}$

Maximum level of virus inactivation detectable (difference between the titre of the viral suspension and the cytotoxicity level):
- Clean conditions ..... $\log 10^{-5.66}$

Reference test (formaldehyde 1.4%)

Cytotoxicity level of formaldehyde 0.7% ..... $\log 10^{-0.50}$

Viral quantification in the reference test (formaldehyde) after 60 minutes and with Poliovirus type 1 ..... $\log 10^{-3.32}$

Viral quantification in the reference test (formaldehyde) after 60 minutes and with Adenovirus type 5 ..... $\log 10^{-1.41}$

Viral quantification in the reference test (formaldehyde) after 60 minutes and with Murine Norovirus ..... $\log 10^{-2,66}$

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Confidence interval

Titre of virus with 95% confidence interval with Poliovirus type 1 (at the requested test time):

- Clean conditions ..... $\log 10^{-7.24 \pm 0.37}$

Titre of virus with 95% confidence interval with Adenovirus type 5 (at the requested test time):

- Clean conditions ..... $\log 10^{-6.33 \pm 0.40}$

Titre of virus with 95% confidence interval with Murine Norovirus (at the requested test time):

- Clean conditions ..... $\log 10^{-6.16 \pm 0.34}$

Reduction with the confidence interval of 95 % .....See table 1.

Sensitivity of cells to virus

- Viral quantification of Poliovirus type 1 with cells not treated by the test solution with the test sample ..... $\log 10^{-7.15}$
- Viral quantification of Poliovirus type 1 with cells treated by the test solution with the test sample ..... $\log 10^{-6.58}$

- Viral quantification of Adenovirus type 5 with cells not treated by the test solution with the test sample ..... $\log 10^{-6.07}$
- Viral quantification of Adenovirus type 5 with cells treated by the test solution with the test sample ..... $\log 10^{-5.66}$

- Viral quantification of Murine Norovirus with cells not treated by the test solution with the test sample ..... $\log 10^{-6.16}$
- Viral quantification of Murine Norovirus with cells treated by the test solution with the test sample ..... $\log 10^{-5.66}$

Note: only can be used to determine the infectivity of cells, those dilutions which: a) show a low degree of cellular destruction (< 25% of cell monolayer) and b) produce a reduction of the title of the virus < 1 $\log_{10}$.

Control of the effectivity of the disinfectant detection activity

- Viral quantification of Poliovirus type 1 after 30 minutes on bath ice without exposing the virus to the test sample ..... $\log 10^{-7.07}$
- Viral quantification of Poliovirus type 1 exposing the virus to the test sample and incubated 30 minutes on ice bath ..... $\log 10^{-6.66}$

- Viral quantification of Adenovirus type 5 after 30 minutes on bath ice without exposing the virus to the test sample ..... $\log 10^{-6.16}$
- Viral quantification of Adenovirus type 5 exposing the virus to the test sample and incubated 30 minutes on ice bath ..... $\log 10^{-5.74}$

- Viral quantification of Murine Norovirus after 30 minutes on bath ice without exposing the virus to the test sample .....$log10^{-6.32}$
- Viral quantification of Murine Norovirus exposing the virus to "the test sample and incubated 30 minutes on ice bath .....$log10^{-5.99}$

Note: The difference between decimal logarithm of titre without exposing the virus to the sample and of the test suspension should be ≤ 0.5

Special remarks

- The sample is tested at 80%; 3% and 0.03%. The highest concentration that can be tested in the test is 80%, because of the mixtures made during the test.

- All controls and validation were between the basic limits.
- One concentration at least showed a log reduction less than 4 log.
- One concentration at least showed a log reduction equal or higher than 4 log.

Assay results

Description of the results under the requested test conditions:

Virus of assay	Test concentrations, reduction obtained with the confidence interval of 95 % and virucidal activity		
	80%	3%	0.03%
Poliovirus type 1	5.33 ± 0.49 $TCID_{50}$ Shows	0.83 ± 0.54 $TCID_{50}$ Does not show	0.17 ± 0.55 $TCID_{50}$ Does not show
Adenovirus type 5	$\geq 5.83 \pm 0.40$ $TCID_{50}$ Shows	1.51 ± 0.52 $TCID_{50}$ Does not show	0.17 ± 0.56 $TCID_{50}$ Does not show
Murine Norovirus	$\geq 5.66 \pm 0.34$ $TCID_{50}$ Shows	2.17 ± 0.48 $TCID_{50}$ Does not show	0.09 ± 0.53 $TCID_{50}$ Does not show

Virucidal activity exists when the titre of virus shows a reduction ≥ 4 log_{10}
$TCID_{50}$: Tissue Culture Infectious Dose 50%.

Tables of results and graphics

See tables 1 to 6 and figure 1 to 3.

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ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 17899/21/ROBCH/Z1

Conclusion

The disinfectant sample **"DEZINFECTANT UNIVERSAL, BIO-DEZ"**, batch not indicated, under clean conditions (bovine serum albumin 0.3 g/L), at **3%** concentration, requested by the client, and during 60 seconds of contact time and 20°C of temperature, **does not show** virucidal activity against the three mandatory viruses (Poliovirus type 1, Adenovirus type 5 and Murine Norovirus) when the activity is assayed according with the NF EN 14476: 2013 + A2: 2019 Standard. However, the disinfectant sample **"DEZINFECTANT UNIVERSAL, BIO-DEZ"**, batch not indicated, under clean conditions (bovine serum albumin 0.3 g/L), at **80%** concentration and during 60 seconds of contact time and 20°C of temperature, **shows** virucidal activity against the three mandatory viruses (Poliovirus type 1, Adenovirus type 5 and Murine Norovirus) when the activity is assayed according with the NF EN 14476: 2013 + A2: 2019 Standard.

Therefore, the disinfectant tested, **does not show general virucidal activity**, diluted at **3%** and **it shows general virucidal activity**, diluted at **80%** when the activity is evaluated according to the NF EN 14476: 2013 + A2: 2019 Standard.

Note 1: The results obtained correspond to the sample received in this laboratory.
Note 2: The information that depend on the information received from the client and are not facilitated by the same one, shown as "not indicated".

Quality Assurance Review:

The assay development and the results obtained have been supervised by the Director of the study.

The Quality Assurance Director has inspected the development of the assay, proving that has been realized following the proper procedure and using the adequate media, materials and reagents, following as well the Good Laboratory Practices (GLPs) and the final report contains the primary data obtained.

Reference:

- **NF EN 14476: 2013 + A2: 2019** Standard. Virucidal quantitative suspension test for chemical disinfectants and antiseptics used in human medicine. Test method and requirements (Phase 2/Step 1). AFNOR.

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ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 17899/21/ROBCH/Z1

Table 1. Results of activity of the sample test sample with Poliovirus type 1 (ATCC VR-192) under test conditions requested by the client.

Assay	Concen-tration*	Interfering substance	Cytoto-xicity level	log_{10} $TCID_{50}$ after…..				Reduction with the confidence interval of 95 %
				0 min	60 sec	30 min	60 min	
Test sample	80%	0.3 g/L BSA	0.5	-	1.91	-	-	5.33 ± 0.49
	3%		0.5	-	6.41	-	-	0.83 ± 0.54
	0.03%		0.5	-	7.07	-	-	0.17 ± 0.55
Virus control	NA	0.3 g/L BSA	NA	7.32	7.24	-	-	NA
Formaldehyde	0.7% (w:v)	NA	0.5	NR	NR	5.16	3.32	NA
Virus control Formaldehyde	0.7% (w:v)	NA	0.5	7.07	NR	NR	6.99	NA

Control of sensitivity of cells to virus (difference between decimal logarithm of titre using treated and untreated cells) ……..….$log10^{-0.57}$

Control of the effectiveness of the disinfectant detection activity (difference between decimal logarithm of titre without exposing the virus to the sample and of the test suspension)…….....$log10^{-0.41}$

NA: not applicable; NR: not realized
Times recommended by Standard for surfaces: maximum 5 or 60 minutes.
Times recommended by Standard for instruments: maximum 60 minutes.
Times recommended by Standard for Hygienic treatment of hands by friction and hygienic handwashing: between 30 or 120 seconds.
PBS: phosphate buffered saline; BSA: bovine serum albumin.
Virucidal activity exists when the titre of virus shows a reduction ≥4 log.
*: see Special remarks to understand the values of these concentrations.

ENCLOSORE NO. 1 SUBCONTRACTED TESTS TO REPORT OF ANALYSIS NO 17899/21/ROBCH/Z1

Table 2. Results of the activity of the test sample, with Poliovirus type 1 (ATCC VR-192) (Assay of titration with 12 wells), under test conditions requested by the client.

Assay	Concentration *	Interfering substance	Time of contact (sec/min)	Dilutions (log10)[a,b] 1	2	3	4	5	6	7	8
Test sample	80%	0.3 g/L BSA	60 sec	4444 4444 4444	2000 3000 0320	0000 0001 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	NR
	3%		60 sec	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	0230 3424 0403	1000 0201 0000	NR
	0.03%		60 sec	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	3042 3343 2444	0201 1302 0020	0000 0000 1100
Cytotoxicity	80%	0.3 g/L BSA	NA	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	NR
Virus control	NA	0.3 g/L BSA	0	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	3023 4040 3320	0000 0102 0000
			60 sec	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	3000 3402 3023	0001 0020 0000
Formaldehyde	0.7% (w:v)	NA	30 min	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	2300 0302 2000	0002 0010 0100	0000 0000 0000	NR
			60 min	4444 4444 4444	4444 4444 4444	0320 2301 0332	0001 0010 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	NR
Control of formaldehyde cytotoxicity	0.7% (w:v)	0.3 g/L BSA	NA	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	NR
Virus control formaldehyde	0.7% (w:v)	NA	0	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	0300 0204 2030	0001 0010 0000
			60 min	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	3240 2444 3403	2301 0100 3202	0000 1000 0000
Sensitivity control of cells to virus	NA	NA	Cells not treated	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC 0CCC 0CCC	0C0C CC0C 0CCC	C000 0000 0000
			Cells treated	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	C0CC CCC0 CCC0	0C00 00C0 000C	0000 000C 0000
Effectiveness control of the disinfectant detection activity	NA	0.3 g/L BSA	Without sample	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	C00C CC0C 0000	0000 00C0 C000
			With sample	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	C0CC CCC0 CCCC	0C00 0C0C C000	0000 0000 0000

a): 1 to 4, virus present and grade of cytophatic effect in 12 units of cellular culture, or grade of cellular lesions in the cytotoxicity assay.
C = cytopathic effect with presence of virus (in this case and according to Standard does not take into account the degree of cytopathic effect only, the presence or absence of the same).
0 = no virus present or absence of cellular lesions in the cytotoxicity assay; NA: not applicable; NR: not realized; BSA: Bovine serum albumin; PBS: phosphate buffered saline.
sec: seconds; min: minutes.*: see Special remarks to understand the values of these concentrations.

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Authorized by: Mariana Ilinca, Manager of Microbiological laboratory
Aprobat de: Mihai Alina-Roxana, General manager (Approved with qualified electronic signature)

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Table 3. Results of activity of the test sample with Adenovirus type 5 (ATCC VR-5), under test conditions requested by the client.

Assay	Concen-tration*	Interfering substance	Cytoto-xicity level	$\log_{10}$ TCID$_{50}$ after…..				Reduction with the confidence interval of 95 %
				0 min	60 sec	30 min	60 min	
Test sample	80%	0.3 g/L BSA	0.5	-	0.50	-	-	≥ 5.83 ± 0.40
	3%		0.5	-	4.82	-	-	1.51 ± 0.52
	0.03%		0.5	-	6.16	-	-	0.17 ± 0.56
Virus control	NA	0.3 g/L BSA	NA	6.41	6.33	-	-	NA
Formaldehyde	0.7% (w:v)	NA	0.5	NR	NR	1.99	1.41	NA
Virus control Formaldehyde	0.7% (w:v)	NA	0.5	6.08	NR	NR	5.98	NA

Control of sensitivity of cells to virus (difference between decimal logarithm of titre using treated and untreated cells) ……..….$\log 10^{-0.41}$

Control of the effectiveness of the disinfectant detection activity (difference between decimal logarithm of titre without exposing the virus to the sample and of the test suspension)……..…$\log 10^{-0.42}$

NA: not applicable; NR: not realized
Times recommended by Standard for surfaces: maximum 5 or 60 minutes
Times recommended by Standard for instruments: maximum 60 minutes
Times recommended by Standard for Hygienic treatment of hands by friction and hygienic handwashing: between 30 or 120 seconds.
PBS: phosphate buffered saline; BSA: bovine serum albumin.
Virucidal activity exists when the titre of virus shows a reduction ≥4 log.
*: see Special remarks to understand the values of these concentrations.

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Table 4. Results of the activity of the test sample, with Adenovirus type 5 (ATCC VR-5) (Assay of titration with 12 wells), under test conditions requested by the client.

Assay	Concentration *	Interfering substance	Time of contact (sec/min)	Dilutions (log10)[a,b] 1	2	3	4	5	6	7	8
Test sample	80%	0.3 g/L BSA	60 sec	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	NR
	3%		60 sec	4444 4444 4444	4444 4444 4444	4444 4444 4444	3342 0324 4344	0200 1202 0002	0000 0000 0000	0000 0000 0000	NR
	0.03%		60 sec	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	0200 0330 2020	0001 0002 0100	NR
Cytotoxicity	80%	0.3 g/L BSA	NA	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	NR
Virus control	NA	0.3 g/L BSA	0	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	3044 3023 4220	0001 0000 2000	NR
			60 sec	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	0200 3304 0332	1000 0020 1000	NR
Formaldehyde	0.7% (w:v)	NA	30 min	4344 2324 4233	0200 1020 0002	0000 0000 1100	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	NR
			60 min	3002 3304 3422	0000 1000 0200	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	NR
Control of formaldehyde cytotoxicity	0.7% (w:v)	0.3 g/L BSA	NA	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	NR
Virus control formaldehyde	0.7% (w:v)	NA	0	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	3404 2430 3043	0201 0311 0220	0010 2000 1000	NR
			60 min	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	3403 4442 3434	0201 0202 0010	0000 0000 1100	NR
Sensitivity control of cells to virus	NA	NA	Cells not treated	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC 0CCC	0C0C CC0C C000	0000 0C00 0C00	NR
			Cells treated	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	C0CC CC0C CCC0	0C00 C0CC 00C0	0000 0000 0000	NR
Effectiveness control of the disinfectant detection activity	NA	0.3 g/L BSA	Without sample	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	C00C C0C0 0CCC	0000 000C 0000	NR
			With sample	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CC00 CCCC	0C00 C0C0 0C00	0000 0000 00C0	NR

a): 1 to 4, virus present and grade of cytophatic effect in 12 units of cellular culture, or grade of cellular lesions in the cytotoxicity assay.
C = cytopathic effect with presence of virus (in this case and according to Standard does not take into account the degree of cytopathic effect only, the presence or absence of the same).
0 = no virus present or absence of cellular lesions in the cytotoxicity assay; NA: not applicable; NR: not realized; BSA: Bovine serum albumin; PBS: phosphate buffered saline.
sec: seconds; min: minutes.
*: see Special remarks to understand the values of these concentrations.

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Table 5. Results of activity of the test sample, with Murine Norovirus, strain S99 Berlin, under test conditions requested by the client.

Assay	Concen-tration*	Interfering substance	Cytoto-xicity level	$\log_{10}$ TCID$_{50}$ after…..				Reduction with the confidence interval of 95 %
				0 min	60 sec	30 min	60 min	
Test sample	80%	0.3 g/L BSA	0.5	-	0.50	-	-	≥5.66 ± 0.34
	3%		0.5	-	3.99	-	-	2.17 ± 0.48
	0.03%		0.5	-	6.07	-	-	0.09 ± 0.53
Virus control	NA	0.3 g/L BSA	NA	6.25	6.16	-	-	NA
Formaldehyde	0.7% (w:v)	NA	0.5	NR	NR	4.24	2.66	NA
Virus control Formaldehyde	0.7% (w:v)	NA	0.5	6.41	NR	NR	6.25	NA

Control of sensitivity of cells to virus (difference between decimal logarithm of titre using treated and untreated cells) ……..…..$\log10^{-0.50}$
0
Control of the effectiveness of the disinfectant detection activity (difference between decimal logarithm of titre without exposing the virus to the sample and of the test suspension)…….…..$\log10^{-0.33}$

NA: not applicable; NR: not realized
Times recommended by Standard for surfaces: maximum 5 or 60 minutes
Times recommended by Standard for instruments: maximum 60 minutes
Times recommended by Standard for Hygienic treatment of hands by friction and hygienic handwashing: between 30 or 120 seconds.
PBS: phosphate buffered saline; BSA: bovine serum albumin.
Virucidal activity exists when the titre of virus shows a reduction ≥4 log.
*: see Special remarks to understand the values of these concentrations.

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Table 6. Results of the activity of the test sample, with Murine Norovirus strain S99 Berlin (Assay of titration with 12 wells), under test conditions requested by the client.

Assay	Concen-tration *	Interfering substance	Time of contact (sec/min)	Dilutions (log10)[a,b]							
				1	2	3	4	5	6	7	8
Test sample	80%	0.3 g/L BSA	60 sec	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	NR
	3%		60 sec	4444 4444 4444	4444 4444 4444	4444 4444 4444	3000 2303 0040	0001 0000 0000	0000 0000 0000	0000 0000 0000	NR
	0.03%		60 sec	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	3443 4203 4434	0200 2012 0302	0011 0000 0000	NR
Cytotoxicity	80%	0.3 g/L BSA	NA	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	NR
Virus control	NA	0.3 g/L BSA	0	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	3444 2334 4434	2011 2201 3201	0000 0000 0000	NR
			60 sec	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	3000 4403 0234	0001 0000 0000	NR
Formaldehyde	0.7% (w:v)	NA	30 min	4444 4444 4444	4444 4444 4444	4444 4444 4444	3430 4203 0220	0001 0000 0000	0000 0000 0000	0000 0000 0000	NR
			60 min	4444 4444 4444	3320 3223 0442	0100 2000 0110	0010 0100 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	NR
Control of formaldehyde cytotoxicity	0.7% (w:v)	0.3 g/L BSA	NA	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	0000 0000 0000	NR
Virus control formaldehyde	0.7% (w:v)	NA	0	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	3004 0320 3330	0010 0002 0021	0000 0000 0000
			60 min	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	4444 4444 4444	0303 0404 3200	0010 0020 1000	0000 0000 0000
Sensitivity control of cells to virus	NA	NA	Cells not treated	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	C000 CC0C 0CC0	000C 00C0 0000	NR
			Cells treated	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CC0C CCCC CCCC	00C0 00CC 0000	0000 0000 0000	NR
Effectiveness control of the disinfectant detection activity	NA	0.3 g/L BSA	Without sample	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	C00C CCC0 CCC0	0000 0C0C 0000	NR
			With sample	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	CCCC CCCC CCCC	C0CC C0CC CCC0	0C00 CC0C 0CCC	00C0 00C0 0000	NR

a): 1 to 4, virus present and grade of cytophatic effect in 12 units of cellular culture, or grade of cellular lesions in the cytotoxicity assay.
C = cytopathic effect with presence of virus (in this case and according to Standard does not take into account the degree of cytopathic effect only, the presence or absence of the same)
0 = no virus present or absence of cellular lesions in the cytotoxicity assay; NA: not applicable; NR: not realized; BSA: Bovine serum albumin; PBS: phosphate buffered saline
sec: seconds; min: minutes
*: see Special remarks to understand the values of these concentrations.

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Figure 1. Results of the activity of the test sample under test conditions requested by the client with Poliovirus type 1 (ATCC VR-192).

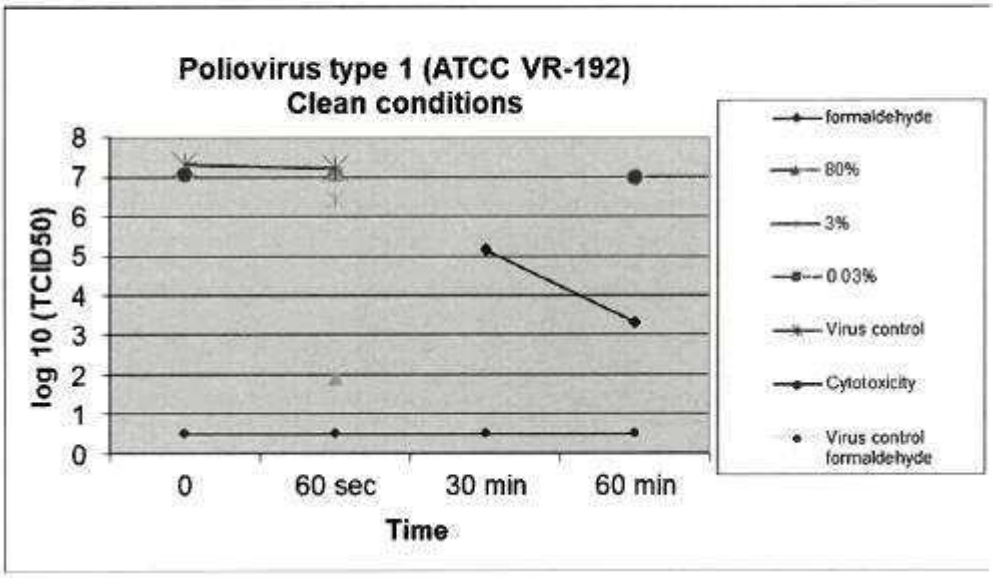

Figure 2. Results of the activity of the test sample under test conditions requested by the client with Adenovirus type 5 (ATCC VR-5).

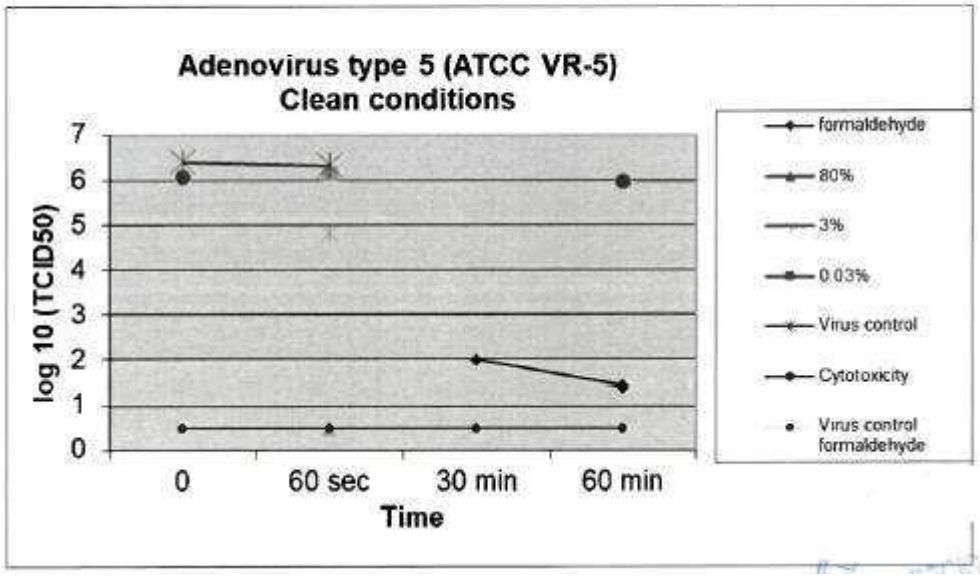

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Figure 3. Results of the activity of the test sample under test conditions requested by the client with Murine Norovirus strain S99 Berlin.