

Test report No. shd1118

EVALUATION OF BACTERICIDAL ACTIVITIES OF A DISINFECTANTS AND ANTISEPTICS USED IN THE MEDICAL AREA (EN 13727)

Name of the product: Chemisept GEL

Batch number: 198251017

Order number: 17030

Manufacturer: Chemi-Pharm Ltd

Client, representative: Chemi-Pharm Ltd., Põllu 132, Tallinn, 10917, ESTONIA
Maris Millner, +372-51-77-090

Date of delivery: 15.01.2018

Test material conditions: No specific features, sample in the manufacturers tare

Storage conditions: In room temperature, dark

Active substance – conc.: Ethyl alcohol – 72.5 % wt; isopropyl alcohol – 7.5 % wt

Appearance of the product: Transparent liquid

Test concentration: Ready to use

Contact time: 15sec, 30 sec

Interfering substance: 15 g/l bovine albumin + 15 ml/l sheep blood erythrocytes =
Dirty conditions; 1,5g/l bovine albumin = clean conditions

Neutralizer: -

Rinsing liquid: Tryptone 1 g/l + NaCl, 9 g/l

Test organisms: *Pseudomonas aeruginosa* ATCC 15442
Staphylococcus aureus ATCC 6538
Enterococcus hirae ATCC 10541
Escherichia coli K12, NTCT 10538
Staphylococcus aureus MRSA ATCC 33592
Enterococcus faecium VRE ATCC 700221

Testing method: EVS-EN 13727:2012+A2:2015
Chemical disinfectants and antiseptics - Quantitative suspension test
for the evaluation of bactericidal activity in the medical area - Test
method and requirements (phase 2, step 1)

Testing date: 17.02.2018 – 19.02.2018

Results: look appendix 1-7



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Diana Kaare, MSc
Head of laboratory, microbiologist
Date of test report: 21.02.2018

Appendix 1

TEST RESULTS (bactericidal suspension test)

EVS-EN 13727:2012+A2:2015; Phase 2, step 1;

Membrane filtration method;

Rinsing liquid: tryptone, 1 g/l+ NaCl, 9 g/l;

Test organism: *Staphylococcus aureus* ATCC 6538;

Test temperature: +20° C; Incubation temperature: +37 °C

Interfering substance: 15 g/l bovine albumin + 15 ml/l sheep blood erythrocytes = Dirty conditions;

1,5g/l bovine albumin = clean conditions

Nordic Tersus Laboratory LLC.;

Date of test: 17.02.2018 – 19.02.2018

Responsible person: Diana Kaare

Validation and controls

Dirty and clean conditions

Validation suspension N_{vo}			Experimental conditions control (A)			Filtration control (B)			Method validation (C)		
V_{C1}	83	$\bar{x} = 86.5$	V_{C1}	72	$\bar{x} = 68.5$	V_{C1}	68	$\bar{x} = 71.5$	V_{C1}	81	$\bar{x} = 77$
V_{C2}	90		V_{C2}	65		V_{C2}	75		V_{C2}	73	
$30 \leq \bar{x} N_{vo} \leq 160$? yes X; no ☐			$\bar{x} A \geq 0,5 \bar{x} N_{vo}$? yes X; no ☐			$\bar{x} B \geq 0,5 \bar{x} N_{vo}$? yes X; no ☐			$\bar{x} C \geq 0,5 \bar{x} N_{vo}$? yes X; no ☐		

Test suspension and test

Testsuspension:	N	V_{C1}	V_{C2}	$\bar{x}_{wm} = 2.45 \times 10^9$; $\log N = 9.39$
	10^{-7}	257	224	$N_0 = N/100$; $\log N_0 = 7,39$
N and N_0	10^{-8}	28	30	$7,17 \leq \log N_0 \leq 7,70$; yes X; no ☐

Experimental results

Concentration of the product %	Dilution step	V_{C1}	V_{C2}	Na ($=\bar{x} \cdot 10$)	log Na	logR	Contact time	Conditions
Ready to use	-	<14	<14	<140	<2.15	>5.24	15 sec	dirty
Ready to use	-	<14	<14	<140	<2.15	>5.24	15 sec	clean
Ready to use	-	<14	<14	<140	<2.15	>5.24	30 sec	dirty
Ready to use	-	<14	<14	<140	<2.15	>5.24	30 sec	clean

Explanations:

V_C = count per ml (one plate or more)

$\bar{x}$ = average of V_{C1} and V_{C2} (1. + 2. duplicate)

N = cfu/ml microbes in testsuspension

N_0 = cfu/ml at the start of the contact time (t=0)

N_{v0} = cfu/ml in the validation suspension (t=0)

Na = surviving microbes after the test

R = reduction factor ($R = N_0 / Na$; $\text{Log}R = \text{Log}N_0 - \text{Log}Na$)

Appendix 2

TEST RESULTS (bactericidal suspension test)

EVS-EN 13727:2012+A2:2015; Phase 2, step 1;
Membrane filtration method;
Rinsing liquid: tryptone, 1 g/l+ NaCl, 9 g/l;
Test organism: *Enterococcus hirae* ATCC 10541;
Test temperature: +20° C; Incubation temperature: +37 °C
Interfering substance: 15 g/l bovine albumin + 15 ml/l sheep blood erythrocytes = Dirty conditions;
1,5g/l bovine albumin = clean conditions
Nordic Tersus Laboratory LLC.;
Date of test: 17.02.2018 – 19.02.2018
Responsible person: Diana Kaare

Validation and controls

Dirty and clean conditions

Validation suspension N_{vo}			Experimental conditions control (A)			Filtration control (B)			Method validation (C)		
V_{C1}	62	$\bar{x} = 66$	V_{C1}	47	$\bar{x} = 46$	V_{C1}	52	$\bar{x} = 53.5$	V_{C1}	59	$\bar{x} = 61$
V_{C2}	70		V_{C2}	45		V_{C2}	55		V_{C2}	53	
$30 \leq \bar{x} N_{vo} \leq 160$? yes X; no ☐			$\bar{x} A \geq 0,5 \bar{x} N_{vo}$? yes X; no ☐			$\bar{x} B \geq 0,5 \bar{x} N_{vo}$? yes X; no ☐			$\bar{x} C \geq 0,5 \bar{x} N_{vo}$? yes X; no ☐		

Test suspension and test

Testsuspension: N and N_0	N	V_{C1}	V_{C2}	$\bar{x}_{wm} = 2.15 \times 10^9$; $\log N = 9.33$ $N_0 = N/100$; $\log N_0 = 7,33$ $7,17 \leq \log N_0 \leq 7,70$; yes X; no ☐
	10^{-7}	203	224	
	10^{-8}	22	25	

Experimental results

Concentration of the product %	Dilution step	V_{C1}	V_{C2}	N_a ($=\bar{x} \cdot 10$)	log N_a	logR	Contact time	Conditions
Ready to use	-	<14	<14	<140	<2.15	>5.18	15 sec	dirty
Ready to use	-	<14	<14	<140	<2.15	>5.18	15 sec	clean
Ready to use	-	<14	<14	<140	<2.15	>5.18	30 sec	dirty
Ready to use	-	<14	<14	<140	<2.15	>5.18	30 sec	clean

Explanations:

V_C = count per ml (one plate or more)

$\bar{x}$ = average of V_{C1} and V_{C2} (1. + 2. duplicate)

N = cfu/ml microbes in testsuspension

N_0 = cfu/ml at the start of the contact time (t=0)

N_{vo} = cfu/ml in the validation suspension (t=0)

N_a = surviving microbes after the test

R = reduction factor ($R = N_0 / N_a$; $\text{Log}R = \text{Log}N_0 - \text{Log}N_a$)

Appendix 3

TEST RESULTS (bactericidal suspension test)

EVS-EN 13727:2012+A2:2015; Phase 2, step 1;

Membrane filtration method;

Rinsing liquid: tryptone, 1 g/l+ NaCl, 9 g/l;

Test organism: *Pseudomonas aeruginosa* ATCC 15442

Test temperature: +20° C; Incubation temperature: +37 °C

Interfering substance: 15 g/l bovine albumin + 15 ml/l sheep blood erythrocytes = Dirty conditions;

1,5g/l bovine albumin = clean conditions

Nordic Tersus Laboratory LLC.;

Date of test: 17.02.2018 – 19.02.2018

Responsible person: Diana Kaare

Validation and controls

Dirty and clean conditions

Validation suspension N_{vo}			Experimental conditions control (A)			Filtration control (B)			Method validation (C)		
V_{C1}	94	$\bar{x} = 87$	V_{C1}	71	$\bar{x} = 72.5$	V_{C1}	66	$\bar{x} = 68$	V_{C1}	75	$\bar{x} = 78.5$
V_{C2}	80		V_{C2}	74		V_{C2}	70		V_{C2}	82	
$30 \leq \bar{x} N_{vo} \leq 160$? yes X; no ☐			$\bar{x} A$ is $\geq 0,5 \bar{x} N_{vo}$? yes X; no ☐			$\bar{x} B$ is $\geq 0,5 \bar{x} N_{vo}$? yes X; no ☐			$\bar{x} C$ is $\geq 0,5 \bar{x} N_{vo}$? yes X; no ☐		

Test suspension and test

Testsuspension:	N	V_{C1}	V_{C2}	$\bar{x}_{wm} = 2.74 \times 10^9$; $\log N = 9.44$
N and N_0	10^{-7}	284	266	$N_0 = N/100$; $\log N_0 = 7,44$
	10^{-8}	24	29	$7,17 \leq \log N_0 \leq 7,70$; yes X; no ☐

Experimental results

Concentration of the product %	Dilution step	V_{C1}	V_{C2}	Na ($=\bar{x} \cdot 10$)	log Na	logR	Contact time	Conditions
Ready to use	-	<14	<14	<140	<2.15	>5.29	15 sec	dirty
Ready to use	-	<14	<14	<140	<2.15	>5.29	15 sec	clean
Ready to use	-	<14	<14	<140	<2.15	>5.29	30 sec	dirty
Ready to use	-	<14	<14	<140	<2.15	>5.29	30 sec	clean

Explanations:

V_C = count per ml (one plate or more)

$\bar{x}$ = average of V_{C1} and V_{C2} (1. + 2. duplicate)

N = cfu/ml microbes in testsuspension

N_0 = cfu/ml at the start of the contact time (t=0)

N_{vo} = cfu/ml in the validation suspension (t=0)

Na = surviving microbes after the test

R = reduction factor ($R = N_0 / Na$; $\text{LogR} = \text{Log}N_0 - \text{Log}Na$)

Appendix 4

TEST RESULTS (bactericidal suspension test)

EVS-EN 13727:2012+A2:2015; Phase 2, step 1;

Membrane filtration method;

Rinsing liquid: tryptone, 1 g/l+ NaCl, 9 g/l;

Test organism: *Escherichia coli* K12, NTCT 10538

Test temperature: +20° C; Incubation temperature: +37 °C

Interfering substance: 15 g/l bovine albumin + 15 ml/l sheep blood erythrocytes = Dirty conditions;

1,5g/l bovine albumin = clean conditions

Nordic Tersus Laboratory LLC.;

Date of test: 17.02.2018 – 19.02.2018

Responsible person: Diana Kaare

Validation and controls

Dirty and clean conditions

Validation suspension N_{vo}			Experimental conditions control (A)			Filtration control (B)			Method validation (C)		
V_{C1}	82	$\bar{x} = 78.5$	V_{C1}	68	$\bar{x} = 60$	V_{C1}	69	$\bar{x} = 69.5$	V_{C1}	70	$\bar{x} = 68.5$
V_{C2}	75		V_{C2}	52		V_{C2}	73		V_{C2}	67	
$30 \leq \bar{x} N_{vo} \leq 160$? yes X; no ☐			$\bar{x} A$ is $\geq 0,5 \bar{x} N_{vo}$? yes X; no ☐			$\bar{x} B$ is $\geq 0,5 \bar{x} N_{vo}$? yesX; no ☐			$\bar{x} C$ is $\geq 0,5 \bar{x} N_{vo}$? yes X; no ☐		

Test suspension and test

Testsuspension: N and N_0	N	V_{C1}	V_{C2}	$\bar{x}_{wm} = 2.48 \times 10^9$; $\log N = 9.39$ $N_0 = N/100$; $\log N_0 = 7.39$ $7,17 \leq \log N_0 \leq 7,70$; yes X; no ☐
	10^{-7}	241	257	
	10^{-8}	22	26	

Experimental results

Concentration of the product %	Dilution step	V_{C1}	V_{C2}	N_a ($=\bar{x} \cdot 10$)	$\log N_a$	$\log R$	Contact time	Conditions
Ready to use	-	<14	<14	<140	<2.15	>5.24	15 sec	dirty
Ready to use	-	<14	<14	<140	<2.15	>5.24	15 sec	clean
Ready to use	-	<14	<14	<140	<2.15	>5.24	30 sec	dirty
Ready to use	-	<14	<14	<140	<2.15	>5.24	30 sec	clean

Explanations:

V_C = count per ml (one plate or more)

$\bar{x}$ = average of V_{C1} and V_{C2} (1. + 2. duplicate)

N = cfu/ml microbes in test suspension

N_0 = cfu/ml at the start of the contact time (t=0)

N_{vo} = cfu/ml in the validation suspension (t=0)

N_a = surviving microbes after the test

R = reduction factor ($R = N_0 / N_a$; $\log R = \log N_0 - \log N_a$)

Appendix5

TEST RESULTS (bactericidal suspension test)

EVS-EN 13727:2012+A2:2015; Phase 2, step 1;

Membrane filtration method;

Rinsing liquid: tryptone, 1 g/l+ NaCl, 9 g/l;

Test organism: *Staphylococcus aureus* MRSA ATCC 33592

Test temperature: +20° C; Incubation temperature: +37 °C

Interfering substance: 15 g/l bovine albumin + 15 ml/l sheep blood erythrocytes = Dirty conditions;

1,5g/l bovine albumin = clean conditions

Nordic Tersus Laboratory LLC.;

Date of test: 17.02.2018 – 19.02.2018

Responsible person: Diana Kaare

Validation and controls

Dirty and clean conditions

Validation suspension N_{vo}			Experimental conditions control (A)			Filtration control (B)			Method validation (C)		
V_{C1}	66	$\bar{x} = 61.5$	V_{C1}	42	$\bar{x} = 45.5$	V_{C1}	49	$\bar{x} = 47.5$	V_{C1}	52	$\bar{x} = 53$
V_{C2}	57		V_{C2}	49		V_{C2}	46		V_{C2}	54	
$30 \leq \bar{x} N_{vo} \leq 160$? yes X; no ☐			$\bar{x} A$ is $\geq 0,5 \bar{x} N_{vo}$? yes X; no ☐			$\bar{x} B$ is $\geq 0,5 \bar{x} N_{vo}$? yes X; no ☐			$\bar{x} C$ is $\geq 0,5 \bar{x} N_{vo}$? yes X; no ☐		

Test suspension and test

Testsuspension:	N	V_{C1}	V_{C2}	$\bar{x}_{wm} = 1.99 \times 10^9$; $\log N = 9.30$
	10^{-7}	204	191	$N_0 = N/100$; $\log N_0 = 7.30$
N and N_0	10^{-8}	18	24	$7,17 \leq \log N_0 \leq 7,70$; yes X; no ☐

Experimental results

Concentration of the product %	Dilution step	V_{C1}	V_{C2}	N_a ($=\bar{x} \cdot 10$)	$\log N_a$	$\log R$	Contact time	Conditions
Ready to use	-	<14	<14	<140	<2.15	>5.15	15 sec	dirty
Ready to use	-	<14	<14	<140	<2.15	>5.15	15 sec	clean
Ready to use	-	<14	<14	<140	<2.15	>5.15	30 sec	dirty
Ready to use	-	<14	<14	<140	<2.15	>5.15	30 sec	clean

Explanations:

V_C = count per ml (one plate or more)

$\bar{x}$ = average of V_{C1} and V_{C2} (1. + 2. duplicate)

N = cfu/ml microbes in testsuspension

N_0 = cfu/ml at the start of the contact time (t=0)

N_{vo} = cfu/ml in the validation suspension (t=0)

N_a = surviving microbes after the test

R = reduction factor ($R = N_0 / N_a$; $\log R = \log N_0 - \log N_a$)

Appendix 6

TEST RESULTS (bactericidal suspension test)

EVS-EN 13727:2012+A2:2015; Phase 2, step 1;

Membrane filtration method;

Rinsing liquid: tryptone, 1 g/l+ NaCl, 9 g/l;

Test organism: *Enterococcus faecium* VRE ATCC 700221

Test temperature: +20° C; Incubation temperature: +37 °C

Interfering substance: 15 g/l bovine albumin + 15 ml/l sheep blood erythrocytes = Dirty conditions;

1,5g/l bovine albumin = clean conditions

Nordic Tersus Laboratory LLC.;

Date of test: 17.02.2018 – 19.02.2018

Responsible person: Diana Kaare

Validation and controls

Dirty and clean conditions

Validation suspension N_{vo}			Experimental conditions control (A)			Filtration control (B)			Method validation (C)		
V_{C1}	62	$\bar{x} = 64$	V_{C1}	52	$\bar{x} = 54.5$	V_{C1}	43	$\bar{x} = 47$	V_{C1}	58	$\bar{x} = 59$
V_{C2}	66		V_{C2}	57		V_{C2}	51		V_{C2}	60	
$30 \leq \bar{x} N_{vo} \leq 160$? yes X; no ☐			$\bar{x} A$ is $\geq 0,5 \bar{x} N_{vo}$? yes X; no ☐			$\bar{x} B$ is $\geq 0,5 \bar{x} N_{vo}$? yes X; no ☐			$\bar{x} C$ is $\geq 0,5 \bar{x} N_{vo}$? yes X; no ☐		

Test suspension and test

Testsuspension:	N	V_{C1}	V_{C2}	$\bar{x}_{wm} = 1.88 \times 10^9$; $\log N = 9.27$
	10^{-7}	181	195	$N_0 = N/100$; $\log N_0 = 7.27$
N and N_0	10^{-8}	22	18	$7,17 \leq \log N_0 \leq 7,70$; yes X; no ☐

Experimental results

Concentration of the product %	Dilution step	V_{C1}	V_{C2}	N_a ($=\bar{x} \cdot 10$)	$\log N_a$	$\log R$	Contact time	Conditions
Ready to use	-	<14	<14	<140	<2.15	>5.12	15 sec	dirty
Ready to use	-	<14	<14	<140	<2.15	>5.12	15 sec	clean
Ready to use	-	<14	<14	<140	<2.15	>5.12	30 sec	dirty
Ready to use	-	<14	<14	<140	<2.15	>5.12	30 sec	clean

Explanations:

V_c = count per ml (one plate or more)

$\bar{x}$ = average of V_{C1} and V_{C2} (1. + 2. duplicate)

N = cfu/ml microbes in test suspension

N_0 = cfu/ml at the start of the contact time (t=0)

N_{vo} = cfu/ml in the validation suspension (t=0)

N_a = surviving microbes after the test

R = reduction factor ($R = N_0 / N_a$; $\log R = \log N_0 - \log N_a$)

Appendix 7

Interpretation:

The product for surgical handrub Chemisept GEL (batch no. 198251017) was tested according to the test method EVS-EN 13727:2012+A2:2015. The test was performed at $20\text{ }^{\circ}\text{C} \pm 1\text{ }^{\circ}\text{C}$, under clean and dirty conditions with the contact times of 15 sec and 30 sec. The membrane filtration method was used for testing the products' effectiveness against the reference strains: *Pseudomonas aeruginosa* ATCC 15442, *Enterococcus hirae* ATCC 10541, *Staphylococcus aureus* ATCC 6538, *Escherichia coli* K12 NTCT 10538, *Staphylococcus aureus* MRSA ATCC 33592 and *Enterococcus faecium* VRE ATCC 700221. Under clean and dirty conditions the tested product was effective against all the reference strains within 15 sec and 30 sec of contact times.

Conclusion:

The surviving count of bacterial reference strains showed at least 5lg reduction meaning that under clean and dirty conditions the ready to use product Chemisept GEL has a bactericidal effect in case of hand disinfection within 15 sec.



Diana Kaare, MSc

Head of laboratory, microbiologist