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Chemical and Microbiological Laboratory, Testing Laboratory No. 1273 certified by Czech Accreditation Institute according to ČSN EN ISO/IEC 17025:2018.

Copy No.: 1
Issue No.: 1

Test report No. S380/2019

DETERMINATION OF VIRUCIDAL (EN 14476:2013 +A2:2019) ACTIVITY OF THE PRODUCT **CHEMISEPT GEL**

Sample ID: S380/2019
Sample name: **Chemisept Gel**
Client: AS CHEMI-PHARM, Pöllu 132, 109 17 Tallinn, Estonia
Producer: AS CHEMI-PHARM, Pöllu 132, 109 17 Tallinn, Estonia
Sampling point: AS CHEMI-PHARM, Pöllu 132, 109 17 Tallinn, Estonia

Incoming date:
9.12.2019

Delivery date:
6.4.2020

Hodonín, 6.4.2020

Ing. Jana Šlitrová, Head of Laboratory

Digitally signed by Moraru Grigore
Date: 2025.11.12 14:06:48 EET
Reason: MoldSign Signature
Location: Moldova
MOLDOVA EUROPEANĂ

Description: *Testing the efficacy of chemical disinfectants and antiseptics*

Sample ID: S380/2019
Rep No: 29
Sample name: **Chemisept Gel**
Sampled: by client
Sampling point: AS CHEMI-PHARM, Pöllu 132, Tallinn, Estonia
Client: AS CHEMI-PHARM, Pöllu 132, 109 17 Tallinn, Estonia

Sampling date: 6.12.2019
Sample delivered: 9.12.2019
Testing date: 4.3. – 13.3.2020
Delivered amount: 100 ml
Batch No: 198031219

Subject of testing:
Determination of virucidal activity of the product.

Identification of the sample:

Name of the product:	**Chemisept Gel**
Batch number:	198031219
Date of manufacture:	03.12.2019
Expiry date:	03.12.2021
Manufacturer:	AS CHEMI-PHARM, Pöllu 132, 109 17 Tallinn, Estonia
Incoming date:	9.12.2019
Storage conditions:	room temperature, dark area
Active ingredients in 100 g:	Ethyl alcohol 72,5 g CAS 64-17-5 Isopropyl alcohol 7,5 g CAS 67-63-0

Experiment conditions:	**Testing of disinfecting efficiency of chemical disinfecting and antiseptic agents by suspension method** SOP-M-19-00 (EN 14476:2013 +A2:2019)
Period of analysis:	4.3. – 13.3.2020
Test temperature:	20 °C ± 1 °C
Method of titration:	virus titration on monolayers of cells on microtitre plates
Appearance of the product:	colourless liquid
Test concentration:	100% (concentrated)*/**/***
Contact time:	1 min, 2 min
Interfering substances:	0.3 g/l BSA (clean conditions)
Reference product:	Formaldehyde 36 – 38% solution p.a., CAS: 50-00-0, Batch No: K51622203930, expiry date: 31.7.2021
Test virus:	*Poliovirus* type 1, LSc-2ab (2nd passage)
Cell lines:	HeLa cells (6th passage)
Incubation:	36 °C ± 1 °C, 5 % CO_2, 96 h, and additional period of 120 hours.

After incubation, the titre infectivity is calculated according to Spearman-Kärber method.

Preparation of the test

1. Determination of the number of the microorganisms CFU/ml in the product
2. Preparation of the cell culture
3. Preparation of the test virus suspension
4. Test of the viral infectivity
5. Virus titration with the interfering substance
6. Cytotoxicity of the product
7. Reference virus inactivation test
8. Test procedure for the virucidal activity of the product

Note:
Virucidal activity – the capability of a product to produce a reduction in the number of infectious virus particles under defined conditions by at least a 4 lg reduction (10^4).
* Product can only be tested at a concentration of 97% (RTU product) or less, as some dilution is always produced by adding the test organisms and interfering substance.
** The test was performed by using MicroSpinTM S 400 HR (2 pcs).
*** The mixture from the product solution and the suspension of virus and the interfering substance makes a clot despite mixing with glass beads.

The standard:
EN 14476:2013 +A2:2019 Chemical disinfectants and antiseptics – Quantitative suspension test for the evaluation of virucidal activity in the medical area – Test method and requirements (Phase 2/Step 1) August 2013 + July 2019

Description: *Testing the efficacy of chemical disinfectants and antiseptics*

Sample ID: S380/2019
Rep No: 29
Sample name: **Chemisept Gel**
Sampled: by client
Sampling point: AS CHEMI-PHARM, Põllu 132, Tallinn, Estonia
Client: AS CHEMI-PHARM, Põllu 132, 109 17 Tallinn, Estonia

Sampling date: 6.12.2019
Sample delivered: 9.12.2019
Testing date: 4.3. – 13.3.2020
Delivered amount: 100 ml
Batch No: 198031219

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

1. Testing the efficacy of chemical disinfectant **Chemisept Gel** on *Poliovirus* type 1, LSc-2ab

Tab No. 1.1 Table of results of product **Chemisept Gel** on *Poliovirus* type 1, LSc-2ab

Product	Concentration **	Interfering substances	Level of cytoxicity	- $\log_{10}$ $TCID_{50}$ after 1 min	- $\log_{10}$ $TCID_{50}$ after 2 min	- $\log_{10}$ $TCID_{50}$ after 30 min	- $\log_{10}$ $TCID_{50}$ after 60 min
Chemisept Gel	100%*/***	clean	≤3.50	5.00	4.50	-	-
Formaldehyde	0.7 % (w/v)	PBS	3.50	-	-	8.67	6.83
			Virus titration, time = 0				
Virus control	-	PBS	10.33	-	-	10.50	10.50
Virus control	-	clean	9.50	-	9.50	-	-

Tab No. 1.2 Testing the efficacy of chemical disinfectant **Chemisept Gel** on *Poliovirus* type 1, LSc-2ab

Test concentration **	Titre of the virus suspension - $\log_{10}$ $TCID_{50}$	Interfering substances	Contact time	- $\log_{10}$ $TCID_{50}$ after test procedure	$\Delta\log_{10}$ $TCID_{50}$
100%*/***	9.50	clean	1 min	5.00	**4.50**
100%*/***	9.50	clean	2 min	4.50	**5.00**

Tab No. 1.3 Testing the efficacy of reference item **Formaldehyde** on *Poliovirus* type 1, LSc-2ab

Test concentration **	Titre of the virus suspension - $\log_{10}$ $TCID_{50}$	Interfering substances	Contact time	- $\log_{10}$ $TCID_{50}$ after test procedure	$\Delta\log_{10}$ $TCID_{50}$
0.7 % (w/v)	10.33	PBS	30 min	8.67	**1.66**
0.7 % (w/v)	10.33	PBS	60 min	6.83	**3.50**

2. Evaluation of virucidal activity of the product **Chemisept Gel**

Tab No. 2.1 The efficacy of chemical disinfectant **Chemisept Gel** on test viruses – virucidal activity

Virucidal activity of the product (EN 14476:2013 +A2:2019)						
Strain	Test temperature [°C]	Contact time [min]	Product test concentrations **	Interfering substances - conditions	$\Delta\log_{10}$ $TCID_{50}$ EN 14476:2013 +A2:2019	$\Delta\log_{10}$ $TCID_{50}$
Poliovirus type 1, LSc-2ab	20	1	100%*/***	clean	≥ 4	**> 4**
Poliovirus type 1, LSc-2ab	20	2	100%*/***	clean	≥ 4	**> 4**

Tab No. 2.2 The efficacy of reference item **Formaldehyde** on test viruses – virucidal activity

Virucidal activity of the product (EN 14476:2013+A2:2019)						
Strain	Test temperature [°C]	Contact time [min]	Product test concentrations **	Interfering substances - conditions	$\Delta\log_{10}$ $TCID_{50}$ EN 14476:2013+ A2:2019	$\Delta\log_{10}$ $TCID_{50}$
Poliovirus type 1, LSc-2ab	20	30	0.7 % (w/v)	PBS	0.5 – 2.5	**1.66**
Poliovirus type 1, LSc-2ab	20	60	0.7 % (w/v)	PBS	2.0 – 4.5	**3.50**

Note:
$TCID_{50}$- 50% infecting dose of a virus suspension or that dilution of the virus suspension that induce a CPE in 50% of cell culture units
* Product can only be tested at a concentration of 97% (RTU product) or less, as some dilution is always produced by adding the test organisms and interfering substance.
** The test was performed by using MicroSpinTM S 400 HR (2 pcs).
*** The mixture from the product solution and the suspension of virus and the interfering substance makes a clot despite mixing with glass beads.
Prepared by: Bc. Iva Čížová, Lab Technician

Description: *Testing the efficacy of chemical disinfectants and antiseptics*

Sample ID: S380/2019
Rep No: 29
Sample name: **Chemisept Gel**
Sampled: by client
Sampling point: AS CHEMI-PHARM, Pöllu 132, Tallinn, Estonia
Client: AS CHEMI-PHARM, Pöllu 132, 109 17 Tallinn, Estonia

Sampling date: 6.12.2019
Sample delivered: 9.12.2019
Testing date: 4.3. – 13.3.2020
Delivered amount: 100 ml
Batch No: 198031219

Interpretation:
Results of tests are in Tabs.
According to EN 14476:2013 +A2:2019 the concentrated*/**/*** tested product **Chemisept Gel,** batch No. 198031219, in the contact times 1 and 2 min under clean conditions at temperature 20 °C ± 1 °C **proved** by the method of virus titration on monolayers of cells on microtitre plates to reduce the number of infectious *Poliovirus* type 1, LSc-2ab, particles under defined conditions by at least a 4 lg reduction.

* Product can only be tested at a concentration of 97% (RTU product) or less, as some dilution is always produced by adding the test organisms and interfering substance.
** The test was performed by using MicroSpinTM S 400 HR (2 pcs).
*** The mixture from the product solution and the suspension of virus and the interfering substance makes a clot despite mixing with glass beads.

Conclusion:
The product **Chemisept Gel** is capable of reducing the number of infectious *Poliovirus* particles under defined conditions (EN 14476:2013 +A2:2019 – concentrated, 1 min and 2 min, clean conditions, 20 °C) to the declared values, and consequently, can be called virucidal on *Poliovirus*.

6.4.2020, Hodonín

Approved by: Ing. Barbora Stoklásková, Leader of Study

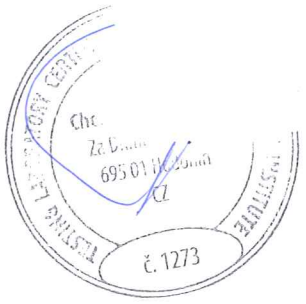
Raw data – product **Chemisept Gel** tested against *Poliovirus* type 1, LSc-2ab
Sample ID: S380/2019, the test report S380/2019,
period of analysis: 4.3. – 13.3.2020
EN14476+A1: *Poliovirus* type 1, LSc-2ab – 2nd passage (NIBSC, GB, 28.3.2018),
HeLa cells – 6th passage (DSMZ, 22.5. 2019)

Appearance of the product: colourless liquid
Test concentration: 100% (concentrated)*/**/***
Contact time: 1 min, 2 min
Interfering substances: 0.3 g/l BSA (clean conditions)
Reference product: Formaldehyde 36 – 38% solution p.a., CAS: 50-00-0, Batch No: K51622203930, expiry date: 31.7.2021

Product	Conc.	Interfering substance	Contact time min	Dilution (lg)[a]									
				2	3	4	5	6	7	8	9	10	11
Chemisept Gel	RTU	clean	1	n.a.	n.a.	333 333	220 200	000 000	000 000	000 000	000 000	000 000	000 000
Chemisept Gel	RTU	clean	2	n.a.	n.a.	333 333	000 000	000 000	000 000	000 000	000 000	000 000	000 000
Chemisept Gel cytotoxicity	RTU	clean	n.a.	n.a.	n.a.	000 000	000 000	000 000	n.d.	n.d.	n.d.	n.d.	n.d.
Chemisept Gel cytotoxicity without Microspin	RTU	clean	n.a.	444 444	444 444	444 444	000 000	000 000	n.d.	n.d.	n.d.	n.d.	n.d.
Formaldehyde	0.7 (w/v)	PBS	30	n.a.	444 444	444 444	333 333	333 333	222 222	220 222	000 220	000 000	000 000
			60	n.a.	444 444	444 333	333 333	200 222	222 020	000 220	000 000	000 000	000 000
Formaldehyde cytotoxicity	0.7 (w/v)	PBS	n.a.	n.a.	444 444	000 000	000 000	000 000	000 000	000 000	000 000	000 000	000 000
Interference control	non-cytotoxic concentration	n.a.	n.a.	n.a.	444 444	444 444	444 444	333 333	333 333	222 222	222 222	222 222	020 000
Neutralization	RTU	clean	n.a.	n.d.	n.d.	444 444	444 444	333 333	333 333	222 222	n.d.	n.d.	n.d.
Virus control	n.a.	PBS	0	n.a.	444 444	444 444	444 444	333 333	333 333	222 222	222 222	222 022	000 000
			30	n.a.	444 444	444 444	444 444	333 333	333 333	222 222	222 222	222 222	000 000
			60	n.a.	444 444	444 444	444 444	333 333	333 333	222 222	222 222	222 222	000 000
Virus control	n.a.	clean	0	n.a.	444 444	444 444	444 444	333 333	333 333	222 222	222 222	000 000	000 000
			2	n.a.	444 444	444 444	444 444	333 333	333 333	222 222	222 222	000 000	000 000
Virus control without Microspin	n.a.	clean	0	444 444	444 444	444 444	444 444	333 333	333 333	222 222	222 222	202 222	000 200

a – dilution, 1 to 4 – degree of CPE in 6 cell culture units, 0 – no CPE
n.a. – not applicable
n.d. – not done
$TCID_{50}$- 50% infecting dose of a virus suspension or that dilution of the virus suspension that induce a CPE in 50% of cell culture units
The calculation of the viral infectivity titre according to EN14476+A2
The formula is:
Negative logarithm of the 50 % end point = negative logarithm of the highest virus concentration used – [({Sum of % affected at each dilution/100} - 0,5) x (lg of dilutions)]
* Product can only be tested at a concentration of 97% (RTU product) or less, as some dilution is always produced by adding the test organisms and interfering substance.
** The test was performed by using MicroSpinTM S 400 HR (2 pcs).
*** The mixture from the product solution and the suspension of virus and the interfering substance makes a clot despite mixing with glass beads.

Prepared by: Bc. Iva Čížová, Lab Technician
Approved by: Ing. Barbora Stoklásková, Leader of Study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