Chemila, spol. s r.o., Za Dráhou 4386/3, Hodonín 69501, Phone +420518340919, chemila@chemila.cz
Chemical and Microbiological Laboratory, Testing Laboratory No. 1273 certified by Czech Accreditation Institute according to ČSN EN ISO/IEC 17025.

Copy No.: 1
Issue No.: 1

Test report No. S37/2018

DETERMINATION OF VIRUCIDAL ACTIVITY (EN 14476+A1) OF THE PRODUCT **MEDI SPRAY**

Sample ID: S37/2018
Sample name: **Medi Spray**
Client: Medi-Sept Sp. z o.o., Konopnica 159c, 210 30 Motycz, Poland
Producer: Medi-Sept Sp. z o.o., Konopnica 159c, 210 30 Motycz, Poland
Sampling point: Medi-Sept Sp. z o.o., Konopnica 159c, 210 30 Motycz, Poland

Incoming date:
27.2.2018

Delivery date:
14.6.2018

Hodonín, 14.6.2018

Ing. Jana Šlitrová, Head of Laboratory

Digitally signed by Palii Nina
Date: 2023.01.04 14:55:17 EET
Reason: MoldSign Signature
Location: Moldova

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

Sample ID: S37/2018
Rep No: 32
Sample name: **Medi Spray**
Sampled: by client
Sampling point: Medi-Sept Sp. z o.o., Konopnica 159c, 210 30 Motycz
Client: Medi-Sept Sp. z o.o., Konopnica 159c, 210 30 Motycz, Poland

Sampling date: 21.2.2018
Sample delivered: 27.2.2018
Testing date: 5.4. – 13.4.2018
Delivered amount: 3 x 100 ml
Batch No: 180123_1

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

Identification of the sample:

Name of the product:	**Medi Spray**
Batch number:	180123_1
Date of manufacture:	2018-01-23
Expiry date:	2021-01-23
Manufacturer:	Medi-Sept Sp. z o.o., Konopnica 159c, 210 30 Motycz, Poland
Incoming date:	27.2.2018
Storage conditions:	at room temperature in dark

Active compounds and concentrations in 100 g:
etanol – 63.7 g
propan-2- ol 6.3 g

Experiment conditions:	**Testing of disinfecting efficiency of chemical disinfecting and antiseptic agents by suspension method** SOP-M-19-00 (EN 14476:2013 +A1:2015)
Period of analysis:	5.4. – 13.4.2018
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:	30 s (0.5 min), 60 s (1 min)
Interfering substances:	0.3 g/l BSA (clean conditions) 3 g/l BSA and 3 ml/l sheep erythrocytes (dirty conditions)
Reference product:	Formaldehyde 36 – 38% solution p.a., CAS: 50-00-0, Batch No: K47740803613, expiry date: 31.3.2018
Test virus:	*BVDV* strain NADL ATCC-VR-534 (6th passage)
Cell lines:	MDBK cells
Incubation:	36 °C ± 1 °C, 5 % CO_2, 96 h, and additional period of 96 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 cell culture
3. Preparation of the test virus suspension
4. Test of viral infectivity
5. Virus titration with interfering substance
6. Cytotoxicity of the product
7. Reference virus inactivation test
8. Test procedure for virucidal activity of 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.
* Product can only be tested at a concentration of 80% or less, as some dilution is always produced by adding the test organisms and interfering substance.
** The test was performed by using MicroSpin™ S 400 HR

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

Sample ID: S37/2018
Rep No: 32
Sample name: **Medi Spray**
Sampled: by client
Sampling point: Medi-Sept Sp. z o.o., Konopnica 159c, 210 30 Motycz
Client: Medi-Sept Sp. z o.o., Konopnica 159c, 210 30 Motycz, Poland

Sampling date: 21.2.2018
Sample delivered: 27.2.2018
Testing date: 5.4. – 13.4.2018
Delivered amount: 3 x 100 ml
Batch No: 180123_1

The standard:
EN 14476:2013 +A1:2015 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 + September 2015

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

1. Testing the efficacy of chemical disinfectant **Medi Spray** on *BVDV* strain NADL ATCC-VR-534

Tab No. 1.1 Table of results of product **Medi Spray** on *BVDV* strain NADL ATCC-VR-534

Product	Concentration**	Interfering substances	Level of cytoxicity	- $\log_{10}$ $TCID_{50}$ after 0.5 min	- $\log_{10}$ $TCID_{50}$ after 1 min	- $\log_{10}$ $TCID_{50}$ after 5 min	- $\log_{10}$ $TCID_{50}$ after 15 min
Medi Spray	100%*	clean	≤2.50	3.50	3.00	-	-
Medi Spray	100%*	dirty	-	3.50	3.00	-	-
Formaldehyde	0.7 % (w/v)	PBS	≤1.50	-	-	6.83	5.67
			Virus titration, time = 0				
Virus control	-	PBS	9.00	9.00	9.00	9.00	8.83
Virus control	-	clean	9.00	9.00	9.00	-	-
Virus control	-	dirty	8.83	8.83	8.83	-	-

Tab No. 1.2 Testing the efficacy of chemical disinfectant **Medi Spray** on *BVDV* strain NADL ATCC-VR-534

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%*	9.00	clean	0.5 min	3.50	**5.50**
100%*	8.83	dirty	0.5 min	3.50	**5.33**
100%*	9.00	clean	1 min	3.00	**6.00**
100%*	8.83	dirty	1 min	3.00	**5.83**

2. Evaluation of virucidal activity of the product **Medi Spray**

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

Virucidal activity of the product (EN 14476:2013+A1:2015)						
Strain	Test temperature [°C]	Contact time [s]	Product test concentrations [%]**	Interfering substances - conditions	$\Delta\log_{10}$ $TCID_{50}$ EN 14476:2013+ A1:2015	$\Delta\log_{10}$ $TCID_{50}$
BVDV strain NADL ATCC-VR-534	20	30	100*	clean	≥ 4	**> 4**
BVDV strain NADL ATCC-VR-534	20	30	100*	dirty	≥ 4	**> 4**
BVDV strain NADL ATCC-VR-534	20	60	100*	clean	≥ 4	**> 4**
BVDV strain NADL ATCC-VR-534	20	60	100*	dirty	≥ 4	**> 4**

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 80% or less, as some dilution is always produced by adding the test organisms and interfering substance.
** The test was performed by using MicroSpin™ S 400 HR

Prepared by: Bc. Iva Čížová, Lab Technician

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

Sample ID: S37/2018
Rep No: 32
Sample name: **Medi Spray**
Sampled: by client
Sampling point: Medi-Sept Sp. z o.o., Konopnica 159c, 210 30 Motycz
Client: Medi-Sept Sp. z o.o., Konopnica 159c, 210 30 Motycz, Poland

Sampling date: 21.2.2018
Sample delivered: 27.2.2018
Testing date: 5.4. – 13.4.2018
Delivered amount: 3 x 100 ml
Batch No: 180123_1

Interpretation:
Results of tests are in Tabs.
The tested concentrated*/** product **Medi Spray**, batch No: 180123_1, in the contact times 30 s and 60 s under clean and dirty 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 *BVDV* strain NADL ATCC-VR-534 particles under defined conditions by at least a 4 lg reduction (EN 14476:2013+A1:2015).

* Product can only be tested at a concentration of 80% or less, as some dilution is always produced by adding the test organisms and interfering substance.
** The test was performed by using MicroSpin™ S 400 HR

Conclusion:
The product **Medi Spray** is capable of reducing the number of infectious *BVDV* particles under defined conditions to the declared values, and consequently, may be called virucidal on *BVDV*.

14.6.2017, Hodonín

.....
Ing. Barbora Stoklásková, Leader of Study

TESTING LABORATORY CERTIFIED BY CZECH ACCREDITATION INSTITUTE
-2-
Chemila, spol. s r.o.
695 01 Hodonín
CZ
Č. 1273

Raw data – product **Medi Spray** tested against *BVDV* strain NADL ATCC-VR-534

Sample S37/2018, the test report S37/2018,

period of analysis: 5.4. – 13.4.2018

EN14476+A1: *BVDV* strain NADL ATCC-VR-534 - 6th passage, MDBK cells – 15th passage

the test conditions: 100%(80%), 30 and 60 s, clean and dirty conditions, 20 °C

Interfering substances: 0.3 g/l BSA (clean conditions)
3 g/l BSA and 3 ml/l sheep erythrocytes (dirty conditions)

Reference product: Formaldehyde 36 – 38% solution p.a., CAS: 50-00-0, Batch No: K47740803613, expiry date: 31.3.2018

Using Microspin

Product	Concentration	Interfering substance	Contact time	Dilution								
				2	3	4	5	6	7	8	9	10
Medi Spray	100%(80%)	clean	30 s	n.a.	222 200	022 000	000 000	000 000	000 000	000 000	000 000	000 000
Medi Spray	100%(80%)	dirty	30 s	n.a.	222 000	000 222	000 000	000 000	000 000	000 000	000 000	000 000
Medi Spray	100%(80%)	clean	60 s	n.a.	022 200	000 000	000 000	000 000	000 000	000 000	000 000	000 000
Medi Spray	100%(80%)	dirty	60 s	n.a.	222 000	000 000	000 000	000 000	000 000	000 000	000 000	000 000
Medi Spray cytotoxicity	100%(80%)	clean	n.a.	n.a.	000 000	000 000	000 000	000 000	n.d.	n.d.	n.d.	n.d.
Formaldehyde K47740803613 Exp: 31.3.2018	0.7 (w/v)	PBS	5 min	444 444	444 444	333 333	222 222	222 202	020 200	000 200	000 000	000 000
			15 min	444 444	444 444	333 333	022 220	220 002	000 000	000 000	000 000	000 000
Formaldehyde cytotoxicity	0.7 (w/v)	PBS	n.a.	000 000	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.	444 444	444 444	444 444	333 333	322 233	222 222	222 222	022 200	000 000
Neutralization	100%(80%)	clean	n.a.	n.d.	n.d.	444 444	333 333	333 333	222 222	222 222	n.d.	n.d.
Virus control	n.a.	PBS	0	444 444	444 444	444 444	333 333	333 333	222 222	222 222	000 222	000 000
			5 min	444 444	444 444	444 444	333 333	323 333	222 222	222 222	200 022	000 000
			15 min	444 444	444 444	444 444	333 333	333 332	222 222	222 222	200 020	000 000
Virus control	n.a.	clean	0	444 444	444 444	444 444	333 333	333 333	222 222	222 222	222 000	000 000
			30 s	444 444	444 444	444 444	333 333	322 333	222 222	222 222	002 220	000 000
			60 s	444 444	444 444	444 444	333 333	333 322	222 222	222 222	000 222	000 000
Virus control	n.a.	dirty	0	444 444	444 444	444 444	333 333	332 333	222 222	222 222	202 000	000 000
			30 s	444 444	444 444	444 444	333 333	323 323	222 222	222 222	002 000	000 200
			60 s	444 444	444 444	444 444	333 333	323 333	222 222	222 222	002 020	000 000

n.a. – not available
n.d. – not done

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