Chemila, Ltd, Hodonín, Blažkova 5, 695 01 Hodonín, CZ, Chemical and Microbiological Laboratory. Laboratory No. 1273 certified by Czech Accreditation Institute. Phone/Fax +420518340919, chemila@iol.cz

Copy No.: 1
Issue No.: 1

Test report No. 1645/2006

Sample ID: 1645/2006
Sample name: **CHLORAMIX DT**
Client: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín
Producer: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín
Sampling point: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín

Incoming date:
11.8.2006

Delivery date:
4.2.2008

Hodonín, 4.2.2008

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Zuzana Matušková, Head of Laboratory

Description: *Testing the efficacy of chemical disinfectants and antiseptics* – SOP-M-19-00

Sample ID: 1645/2006
Rep No: 48
Sample name: **CHLORAMIX DT**
Sampled: by client
Sampling point: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín
Client: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín

Sampling date: 11.8.2006
Sample delivered: 11.8.2006
Testing date: 22.8.2006-8.11.2006
Delivered amount: 500 g
Batch No: 06-018-0-LO-00:29

Subject of testing:
Determination of bactericidal and fungicidal activity - dilution neutralization method and on carriers, tuberculocidal and mycobactericidal activity, virucidal activity of the product.

Identification of the sample:

Name of the product:	**CHLORAMIX DT**
Batch number:	06-018-0-LO-00:29
Date of manufacture:	01/04
Expiry date:	01/09
Manufacturer:	BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín
Incoming date:	11.8.2006
Storage conditions:	stated by the manufacturer
Active compouds and concentrations:	CAS 51580-86-0 sodium dichlorisocyanurate

Experimental conditions:

	Quantitative suspension test for evaluation of bactericidal activity SOP-M-19-00-A (ČSN EN 13727, ČSN EN 1276)
Period of analysis:	22.8.2006-23.8.2006
Test temperature:	20 °C ± 1 °C
Test method:	dilution neutralization method
Neutralization medium:	Dey-Engley Neutralizing Broth M 1062
Appearance of the products:	white tablets
Product diluent:	hard water
Test concentration:	1 tablet/ 5 l (3.33 g/5 l = 0.66 g/l, 0.066%)
Contact time:	15 min
Interfering substances:	3 g/l BSA and 3 ml/l sheep erythrocytes (dirty conditions)
Test organisms:	*Escherichia coli* CCM 3954
	Pseudomonas aeruginosa CCM 3955
	Staphylococcus aureus CCM 3953
	Enterococcus hirae CCM 2423

Test procedure:
1. Preparation of test suspension
2. Counting of test suspension
3. Quantitative suspension test

Note:
Bactericidal activity – the capability of a product to produce a reduction in the number of viable bacterial cells of relevant organisms under defined conditions by at least 5 orders (10^5).
$\Delta \log N = \log N_{\text{cfu/ml test suspension}} - \log N_{\text{cfu/ml after test procedure}}$

The standard:
ČSN EN 13727 Chemical disinfectants and antiseptic – Quantitative suspension test for evaluation of bactericidal activity for instruments used in the medical area - Test method and requirements (phase 2, step 1),
ČSN EN 1276 Chemical disinfectants and antiseptic – Quantitative suspension test for the evaluation of bactericidal activity of chemical disinfectants and aniseptics used in food, industrial, domestic, and institutional area - Test method and requirements (phase 2, step 1)

Description: *Testing the efficacy of chemical disinfectants and antiseptics* – SOP-M-19-00

Sample ID: 1645/2006
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The Number of CFU in the tested product **CHLORAMIX DT**: 0 CFU/ml

Tab No. 1 Testing the efficacy of chemical disinfectant **CHLORAMIX DT** on *Escherichia coli*

Test concentration (g/l) / contact time (min) / conditions	Dilution after test procedure	Count CFU/ml	N	log N	Δlog N
0.66/15/dirty	10^{-2}	0	0	<2	≥7.67
Test suspension	10^{-8}	47	$4.7 \cdot 10^{9}$	9.67	

Tab No. 2 Testing the efficacy of chemical disinfectant **CHLORAMIX DT** on *Pseudomonas aeruginosa*

Test concentration (g/l) / contact time (min) / conditions	Dilution after test procedure	Count CFU/ml	N	log N	Δlog N
0.66/15/dirty	10^{-2}	0	0	<2	≥7.64
Test suspension	10^{-8}	44	$4.4 \cdot 10^{9}$	9.64	

Tab No. 3 Testing the efficacy of chemical disinfectant **CHLORAMIX DT** on *Staphylococcus aureus*

Test concentration (g/l) / contact time (min) / conditions	Dilution after test procedure	Count CFU/ml	N	log N	Δlog N
0.66/15/dirty	10^{-2}	0	0	<2	≥7.98
Test suspension	10^{-8}	96	$9.6 \cdot 10^{9}$	9.98	

Tab No. 4 Testing the efficacy of chemical disinfectant **CHLORAMIX DT** on *Enterococcus hirae*

Test concentration (g/l) / contact time (min) / conditions	Dilution after test procedure	Count CFU/ml	N	log N	Δlog N
0.66/15/dirty	10^{-3}	52	$5.2 \cdot 10^{4}$	4.72	5.24
Test suspension	10^{-8}	91	$9.1 \cdot 10^{9}$	9.96	

$\Delta\log N = \log N_{\text{cfu/ml test suspension}} - \log N_{\text{cfu/ml after test procedure}}$

Tab No. 5 The efficacy of chemical disinfectant **CHLORAMIX DT** on test strains – bactericidal activity

Bactericidal activity of the product (ČSN EN 13727, ČSN EN 1276)							
Strain	T [°C]	Contact time [min]	Product test concentrations [g/l (%)]	Interfering substances - conditions	Δlog N ČSN EN 13727	Δlog N ČSN EN 1276	**Δlog N**
Escherichia coli	20	15	0.66 (0.066)	dirty	> 5	> 5	> 5
Pseudomonas aeruginosa	20	15	0.66 (0.066)	dirty	> 5	> 5	> 5
Staphylococcus aureus	20	15	0.66 (0.066)	dirty	> 5	> 5	> 5
Enterococcus hirae	20	15	0.66 (0.066)	dirty	> 5	> 5	> 5

Prepared by: Hana Konevalíková, Lab Assistant

Description: *Testing the efficacy of chemical disinfectants and antiseptics* – SOP-M-19-00

Sample ID: 1645/2006
Rep No: 48
Sample name: **CHLORAMIX DT**
Sampled: by client
Sampling point: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín
Client: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín

Sampling date: 11.8.2006
Sample delivered: 11.8.2006
Testing date: 22.8.2006-8.11.2006
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Experimental conditions:

Quantitative suspension test for evaluation of fungicidal activity
SOP-M-19-00-B (ČSN EN 13624, ČSN EN 1650)

Period of analysis:	23.8.2006-28.8.2006
Test temperature:	20 °C ± 1 °C
Test method:	dilution neutralization method
Neutralization medium:	Dey-Engley Neutralizing Broth M 1062
Appearance of the products:	white tablets
Product diluent:	hard water
Test concentration:	1 tablet/ 5 l (3.33 g/5 l = 0.66 g/l, 0.066%)
Test concentration:	1 tablet/ 3 l (3.33 g/3 l = 1.11 g/l, 0.111%)
Contact time:	15 min
Interfering substances:	3 g/l BSA and 3 ml/l sheep erythrocytes (dirty conditions)
Test organisms:	*Candida albicans* CCM 8186
	Aspergillus niger CCM 8222

Test procedure:

1. Preparation of test suspension
2. Counting of test suspension
3. Quantitative suspension test

Note:
Fungicidal activity – the capability of a product to produce at least a 10^4 reduction in the number of viable vegetative yeast cells and mould spores belonging to reference strains of relevant organisms under defined conditions.
$\Delta \log N = \log N_{\text{cfu/ml test suspension}} - \log N_{\text{cfu/ml after test procedure}}$

The standard:
ČSN EN 13624 Chemical disinfectants and antiseptic - Quantitative suspension test for evaluation of fungicidal activity of chemical disinfectants for instruments used in the medical area - Test method and requirements (Phase 2/Step 1)
ČSN EN 1650 Chemical disinfectants and antiseptic - Quantitative suspension test for the evaluation of fungicidal activity of chemical disinfectants used in food, industrial, domestic, and institutional area - Test method and requirements (phase 2, step 1)

Description: *Testing the efficacy of chemical disinfectants and antiseptics* – SOP-M-19-00

Sample ID: 1645/2006
Rep No: 48
Sample name: **CHLORAMIX DT**
Sampled: by client
Sampling point: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín
Client: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín

Sampling date: 11.8.2006
Sample delivered: 11.8.2006
Testing date: 22.8.2006-8.11.2006
Delivered amount: 500 g
Batch No: 06-018-0-LO-00:29
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Tab No. 6 Testing the efficacy of chemical disinfectant **CHLORAMIX DT** on *Candida albicans*

Test concentration (g/l) / contact time (min) / conditions	Dilution after test procedure	Count CFU/ml	N	log N	Δlog N
0.66/15/dirty	10^{-2}	0	0	<2	≥7.15
1.11/15/dirty	10^{-2}	0	0	<2	≥7.15
Test suspension	10^{-7}	141	$1.41 \cdot 10^{9}$	9.15	

Tab No. 7 Testing the efficacy of chemical disinfectant **CHLORAMIX DT** on *Aspergillus niger*

Test concentration (g/l) / contact time (min) / conditions	Dilution after test procedure	Count CFU/ml	N	log N	Δlog N
0.66/15/dirty	10^{-2}	0	0	<2	≥7.75
1.11/15/dirty	10^{-2}	0	0	<2	≥7.75
Test suspension	10^{-8}	56	$5.6 \cdot 10^{9}$	9.75	

$\Delta\log N = \log N_{\text{cfu/ml test suspension}} - \log N_{\text{cfu/ml after test procedure}}$

Tab No. 8 The efficacy of chemical disinfectant **CHLORAMIX DT** on test strains – fungicidal activity

Fungicidal activity of the product (ČSN EN 13624, ČSN EN 1650)							
Strain	T [°C]	Contact time [min]	Product test concentrations [g/l (%)]	Interfering substances - conditions	Δlog N ČSN EN 13624	Δlog N ČSNEN 1650	Δlog N
Candida albicans	20	15	0.66 (0.066)	dirty	> 4	> 4	> 4
Aspergillus niger	20	15	0.66 (0.066)	dirty	> 4	> 4	> 4
Candida albicans	20	15	1.11 (0.111)	dirty	> 4	> 4	> 4
Aspergillus niger	20	15	1.11 (0.111)	dirty	> 4	> 4	> 4

Prepared by: Hana Konevalíková, Lab Assistant

Description: *Testing the efficacy of chemical disinfectants and antiseptics* – SOP-M-19-00

Sample ID: 1645/2006
Rep No: 48
Sample name: **CHLORAMIX DT**
Sampled: by client
Sampling point: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín
Client: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín

Sampling date: 11.8.2006
Sample delivered: 11.8.2006
Testing date: 22.8.2006-8.11.2006
Delivered amount: 500 g
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Experimental conditions:

Quantitative surface test for evaluation of bactericidal activity and fungicidal activity
SOP-M-19-00-C (ČSN EN 13697)

Period of analysis:	11.10.2006-12.10.2006 (bact.), 11.10.2006-16.10.2006 (fung.)
Test temperature:	20 °C ± 1 °C
Test method:	dilution neutralization method
Neutralization medium:	Dey-Engley Neutralizing Broth M 1062
Appearance of the products:	white tablets
Product diluent:	hard water
Test concentration:	1 tablet/ 5 l (3.33 g/5 l = 0.66 g/l, 0.066%)
Contact time:	15 min
Interfering substances:	3 g/l BSA and 3 ml/l sheep erythrocytes (dirty conditions)
Test concentration:	1 tablet/ 10 l (3.33 g/10 l = 0.33 g/l, 0.033%)
Contact time:	15 min
Interfering substances:	0.3 g/l BSA (clean conditions)
Test organisms:	*Escherichia coli* CCM 3954
	Pseudomonas aeruginosa CCM 3955
	Staphylococcus aureus CCM 3953
	Enterococcus hirae CCM 2423
	Candida albicans CCM 8186
	Aspergillus niger CCM 8222

Test procedure:

1. Preparation of test suspension
2. Counting of test suspension
3. Quantitative surface test

Note:
Bactericidal activity – the capability of a product to produce at least 10^4 reduction in the number of viable bacterial cells belonging to reference strains under defined conditions.
Fungicidal activity – the capability of a product to produce at least 10^3 reduction in the number of viable fungi belonging to reference strains under defined conditions.
$\Delta \log N = \log N_{\text{cfu/ml test suspension}} - \log N_{\text{cfu/ml after test procedure}}$

The standard:
ČSN EN 13697 Chemical disinfectants and antiseptic – Quantitative non-porous test for evaluation of bactericidal and/or fungicidal activity of chemical disinfectants used in food, industrial, domestic, and institutional area - Test method and requirements without mechanical action (phase 2/ step 2))

Description: *Testing the efficacy of chemical disinfectants and antiseptics* – SOP-M-19-00

Sample ID: 1645/2006
Rep No: 48
Sample name: **CHLORAMIX DT**
Sampled: by client
Sampling point: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín
Client: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín

Sampling date: 11.8.2006
Sample delivered: 11.8.2006
Testing date: 22.8.2006-8.11.2006
Delivered amount: 500 g
Batch No: 06-018-0-LO-00:29

Tab No. 9 Testing the efficacy of chemical disinfectant **CHLORAMIX DT** on *Escherichia coli* on carrier

Test concentration (g/l) / contact time (min) / conditions	Dilution after test procedure	Count CFU/ml	N	log N	Δlog N
0.66/15/dirty	10^{-3}	0	0	<3	≥5.26
0.33/15/clean	10^{-3}	0	0	<3	≥5.26
Test suspension	10^{-7}	18	$1.8 \cdot 10^{8}$	8.26	

Tab No. 10 Testing the efficacy of chemical disinfectant **CHLORAMIX DT** on *Pseudomonas aeruginosa* on carrier

Test concentration (g/l) / contact time (min) / conditions	Dilution after test procedure	Count CFU/ml	N	log N	Δlog N
0.66/15/dirty	10^{-3}	0	0	<3	≥5.71
0.33/15/clean	10^{-3}	0	0	<3	≥5.71
Test suspension	10^{-7}	51	$5.1 \cdot 10^{8}$	8.71	

Tab No. 11 Testing the efficacy of chemical disinfectant **CHLORAMIX DT** on *Staphylococcus aureus* on carrier

Test concentration (g/l) / contact time (min) / conditions	Dilution after test procedure	Count CFU/ml	N	log N	Δlog N
0.66/15/dirty	10^{-3}	0	0	<3	≥5.53
0.33/15/clean	10^{-3}	0	0	<3	≥5.53
Test suspension	10^{-7}	34	$3.4 \cdot 10^{8}$	8.53	

Tab No. 12 Testing the efficacy of chemical disinfectant **CHLORAMIX DT** on *Enterococcus hirae* on carrier

Test concentration (g/l) / contact time (min) / conditions	Dilution after test procedure	Count CFU/ml	N	log N	Δlog N
0.66/15/dirty	10^{-3}	0	0	<3	≥5.18
0.33/15/clean	10^{-3}	0	0	<3	≥5.18
Test suspension	10^{-7}	15	$1.5 \cdot 10^{8}$	8.18	

$\Delta \log N = \log N_{\text{cfu/ml test suspension}} - \log N_{\text{cfu/ml after test procedure}}$

Description: *Testing the efficacy of chemical disinfectants and antiseptics* – SOP-M-19-00

Sample ID: 1645/2006
Rep No: 48
Sample name: **CHLORAMIX DT**
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Sampling point: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín
Client: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín

Sampling date: 11.8.2006
Sample delivered: 11.8.2006
Testing date: 22.8.2006-8.11.2006
Delivered amount: 500 g
Batch No: 06-018-0-LO-00:29

Tab No. 13 The efficacy of chemical disinfectant **CHLORAMIX DT** on bacterial strains – bactericidal activity on carrier

Bactericidal activity of the product on carrier (ČSN EN 13697)						
Strain	Test temperature [°C]	Contact time [min]	Product test concentrations [g/l (%)]	Interfering substances - conditions	ČSN EN 13697	Δlog N
Escherichia coli	20	15	0.33 (0.033)	clean	> 4	> 4
Pseudomonas aeruginosa	20	15	0.33 (0.033)	clean	> 4	> 4
Staphylococcus aureus	20	15	0.33 (0.033)	clean	> 4	> 4
Enterococcus hirae	20	15	0.33 (0.033)	clean	> 4	> 4
Escherichia coli	20	15	0.66 (0.066)	dirty	> 4	> 4
Pseudomonas aeruginosa	20	15	0.66 (0.066)	dirty	> 4	> 4
Staphylococcus aureus	20	15	0.66 (0.066)	dirty	> 4	> 4
Enterococcus hirae	20	15	0.66 (0.066)	dirty	> 4	> 4

Tab No. 14 Testing the efficacy of chemical disinfectant **CHLORAMIX DT** on *Candida albicans* on carrier

Test concentration (g/l) / contact time (min) / conditions	Dilution after test procedure	Count CFU/ml	N	log N	Δlog N
0.66/15/dirty	10^{-3}	0	0	<3	≥5.20
0.33/15/clean	10^{-3}	0	0	<3	≥5.20
Test suspension	10^{-7}	16	$1.6 \cdot 10^{8}$	8.20	

Tab No. 15 Testing the efficacy of chemical disinfectant **CHLORAMIX DT** on *Aspergillus niger* on carrier

Test concentration (g/l) / contact time (min) / conditions	Dilution after test procedure	Count CFU/ml	N	log N	Δlog N
0.66/15/dirty	10^{-3}	0	0	<3	≥5.32
0.33/15/clean	10^{-3}	0	0	<3	≥5.32
Test suspension	10^{-7}	21	$2.1 \cdot 10^{8}$	8.32	

$\Delta\log N = \log N_{\text{cfu/ml test suspension}} - \log N_{\text{cfu/ml after test procedure}}$

Tab No. 16 The efficacy of chemical disinfectant **CHLORAMIX DT** on fungal strains – fungicidal activity on carrier

Fungicidal activity of the product on carrier (ČSN EN 13697)						
Strain	Test temperature [°C]	Contact time [min]	Product test concentrations [ml/l (%)]	Interfering substances - conditions	ČSN EN 13697	Δlog N
Candida albicans	20	15	0.33 (0.033)	clean	> 3	> 3
Aspergillus niger	20	15	0.33 (0.033)	clean	> 3	> 3
Candida albicans	20	15	0.66 (0.066)	dirty	> 3	> 3
Aspergillus niger	20	15	0.66 (0.066)	dirty	> 3	> 3

Prepared by: Hana Konevalíková, Lab Assistant

Description: *Testing the efficacy of chemical disinfectants and antiseptics* – SOP-M-19-00

Sample ID: 1645/2006
Rep No: 48
Sample name: **CHLORAMIX DT**
Sampled: by client
Sampling point: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín
Client: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín

Sampling date: 11.8.2006
Sample delivered: 11.8.2006
Testing date: 22.8.2006-8.11.2006
Delivered amount: 500 g
Batch No: 06-018-0-LO-00:29

Experiment conditions: **Quantitative test for evaluation of virucidal activity**
SOP-M-19-00-H (ČSN EN 14476)

Period of analysis:	19.9.2006-30.9.2006
Test temperature:	20 °C ± 1 °C
Method of titration:	virus titration on monolayers of cells in cell culture tubes
Appearance of the products:	white tablets
Product diluent:	hard water
Test concentration:	1 tablet/ 1.5 l (3.33 g/1.5 l = 2.22 g/l, 0.222%)
Contact time:	15 min
Interfering substances:	3 g/l BSA and 3 ml/l sheep erythrocytes (dirty conditions)
Test concentration:	1 tablet/ 5 l (3.33 g/5 l = 0.66 g/l, 0.066%)
Contact time:	15 min
Interfering substances:	0.3 g/l BSA (clean conditions) 3 g/l BSA and 3 ml/l sheep erythrocytes (dirty conditions)

Procedure to stop action of product: The virucidal activity is immediately suppressed by transfer of the sample into 9 volumes of ice-cold diluent.

Reference product:	Formaldehyde 36 – 38%solution p.a., CAS: 50-00-0, Batch No: 232-08052006, expiry date: 05.2008
Test virus:	*poliovirus* type 1, LSc-2ab
Cell lines:	HeLa cells

Titre values are calculated according to Spaerman and Kärber.

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 4 orders (10^4).

The standard:
ČSN EN 14476 Chemical disinfectants and antiseptics – Virucidal quantitative test for chemical disinfectants and antiseptics – Test method and requirements (phase 2/step 1)

Description: *Testing the efficacy of chemical disinfectants and antiseptics* – SOP-M-19-00

Sample ID: 1645/2006
Rep No: 48
Sample name: **CHLORAMIX DT**
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Sampling point: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín
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Sampling date: 11.8.2006
Sample delivered: 11.8.2006
Testing date: 22.8.2006-8.11.2006
Delivered amount: 500 g
Batch No: 06-018-0-LO-00:29
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Experiment conditions:	**Quantitative test for evaluation of virucidal activity** SOP-M-19-00-H (ČSN EN 14476)
Period of analysis:	6.10.2006-16.10.2006
Test temperature:	20 °C ± 1 °C
Method of titration:	virus titration on monolayers of cells in cell culture tubes
Appearance of the products:	white tablets
Product diluent:	hard water
Test concentration:	1 tablet/ 1.5 l (3.33 g/1.5 l = 2.22 g/l, 0.222%)
Contact time:	15 min
Interfering substances:	3 g/l BSA and 3 ml/l sheep erythrocytes (dirty conditions)
Test concentration:	1 tablet/ 5 l (3.33 g/5 l = 0.66 g/l, 0.066%)
Contact time:	15 min
Interfering substances:	0.3 g/l BSA (clean conditions) 3 g/l BSA and 3 ml/l sheep erythrocytes (dirty conditions)

Procedure to stop action of product: The virucidal activity is immediately suppressed by transfer of the sample into 9 volumes of ice-cold diluent.

Test virus:	*adenovirus* type 5
Cell lines:	HeLa cells

Titre values are calculated according to Spaerman and Kärber.

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. 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 4 orders (10^4).

The standard:
ČSN EN 14476 Chemical disinfectants and antiseptics – Virucidal quantitative test for chemical disinfectants and antiseptics – Test method and requirements (phase 2/step 1)

Description: *Testing the efficacy of chemical disinfectants and antiseptics* – SOP-M-19-00

Sample ID: 1645/2006
Rep No: 48
Sample name: **CHLORAMIX DT**
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Delivered amount: 500 g
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Experiment conditions:	**Quantitative test for evaluation of virucidal activity** SOP-M-19-00-H
Period of analysis:	29.9.2006-9.10.2006
Test temperature:	20 °C ± 1 °C
Method of titration:	virus titration on monolayers of cells in cell culture tubes
Appearance of the products:	white tablets
Diluent:	hard water
Test concentration:	1 tablet/ 1.5 l (3.33 g/1.5 l = 2.22 g/l, 0.222%)
Contact time:	15 min
Interfering substances:	3 g/l BSA and 3 ml/l sheep erythrocytes (dirty conditions)

Procedure to stop action of product: The virucidal activity is immediately suppressed by transfer of the sample into 9 volumes of ice-cold diluent.

Test virus:	BVDV strain NADL ATCC-VR-534
Cell lines:	MDBK cells

Titre values are calculated according to Spaerman and Kärber.

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. 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 4 orders (10^4).

Description: *Testing the efficacy of chemical disinfectants and antiseptics* – SOP-M-19-00

Sample ID: 1645/2006
Rep No: 48
Sample name: **CHLORAMIX DT**
Sampled: by client
Sampling point: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín
Client: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín

Sampling date: 11.8.2006
Sample delivered: 11.8.2006
Testing date: 22.8.2006-8.11.2006
Delivered amount: 500 g
Batch No: 06-018-0-LO-00:29

Tab No. 17 Table of results of product **CHLORAMIX DT** on *poliovirus* type 1, LSc-2ab

Product	Concentration	Interfering substances	Level of cyttoxicity	- $\log_{10}ID_{50}$ after 15 minutes	- $\log_{10}ID_{50}$ after 30 minutes
CHLORAMIX DT	0.222%	with 3 g/l BSA	2.9	3.3	-
CHLORAMIX DT	0.066%	with 3 g/l BSA	2.1	6.7	-
CHLORAMIX DT	0.066%	with 0.3 g/l BSA	2.1	2.7	-
Formaldehyde	0.7 % (w/v)	PBS	3.5	-	8.2
Virus control	-	PBS	-	-	10.1
Virus control	-	with 3 g/l BSA	-	10.3	-
Virus control	-	with 0.3 g/l BSA	-	10.1	-

Tab No. 18 Testing the efficacy of chemical disinfectant **CHLORAMIX DT** on *poliovirus* type 1, LSc-2ab

Test concentration	Titre of the virus suspension - $\log_{10}ID_{50}$	Interfering substances	Contact time	- $\log_{10}ID_{50}$ after test procedure	$\Delta\log_{10}ID_{50}$
0.222%	10.3	with 3 g/l BSA	15 min	3.3	**7.0**
0.066%	10.3	with 3 g/l BSA	15 min	6.7	**3.6**
0.066%	10.1	with 0.3 g/l BSA	15 min	2.7	**7.4**

Prepared by: Hana Konevalíková, Lab Assistant

Tab No. 19 Table of results of product **CHLORAMIX DT** on *adenovirus* type 5

Product	Concentration	Interfering substances	Level of cyttoxicity	$\log_{10}ID_{50}$ after 15 minutes
CHLORAMIX DT	0.222%	with 3 g/l BSA	2.9	6.5
CHLORAMIX DT	0.066%	with 3 g/l BSA	2.1	7.7
CHLORAMIX DT	0.066%	with 0.3 g/l BSA	2.1	6.1
Virus control	-	with 3 g/l BSA	-	10.1
Virus control	-	with 0.3 g/l BSA	-	10.1

Tab No. 20 Testing the efficacy of chemical disinfectant **CHLORAMIX DT** *adenovirus* type 5

Test concentration	Titre of the virus suspension - $\log_{10}ID_{50}$	Interfering substances	Contact time	- $\log_{10}ID_{50}$ after test procedure	$\Delta\log_{10}ID_{50}$
0.222%	10.1	with 3 g/l BSA	15 min	6.5	**3.6**
0.066%	10.1	with 3 g/l BSA	15 min	7.7	**2.4**
0.066%	10.1	with 0.3 g/l BSA	15 min	6.1	**4.0**

Prepared by: Hana Konevalíková, Lab Assistant

Description: *Testing the efficacy of chemical disinfectants and antiseptics* – SOP-M-19-00

Sample ID: 1645/2006
Rep No: 48
Sample name: **CHLORAMIX DT**
Sampled: by client
Sampling point: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín
Client: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín

Sampling date: 11.8.2006
Sample delivered: 11.8.2006
Testing date: 22.8.2006-8.11.2006
Delivered amount: 500 g
Batch No: 06-018-0-LO-00:29

Tab No. 21 Table of results of product **CHLORAMIX DT** on on virus BVDV strain NADL

Product	Concentration	Interfering substances	Level of cyttoxicity	$log_{10}ID_{50}$ after 15 minutes
CHLORAMIX DT	0.222%	with 3 g/l BSA	3.1	4.7
Virus control	-	with 3 g/l BSA	-	9.1

Tab No. 22 Testing the efficacy of chemical disinfectant **CHLORAMIX DT** on virus BVDV strain NADL

Test concentration	Titre of the virus suspension - $log_{10}ID_{50}$	Interfering substances	Contact time	- $log_{10}ID_{50}$ after test procedure	$\Delta log_{10}ID_{50}$
0.222%	9.1	with 3 g/l BSA	15 min	4.7	**4.4**

Prepared by: Iva Čížová, Lab Assistant

Description: *Testing the efficacy of chemical disinfectants and antiseptics* – SOP-M-19-00

Sample ID: 1645/2006
Rep No: 48
Sample name: **CHLORAMIX DT**
Sampled: by client
Sampling point: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín
Client: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín

Sampling date: 11.8.2006
Sample delivered: 11.8.2006
Testing date: 22.8.2006-8.11.2006
Delivered amount: 500 g
Batch No: 06-018-0-LO-00:29
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Experimental conditions:

	Quantitative suspension test for evaluation of mycobactericidal activity SOP-M-19-00-D (ČSN EN 14348)
Period of analysis:	12.10.2006-8.11.2006
Test temperature:	20 °C ± 1 °C
Test method:	dilution neutralization method
Neutralization medium:	Dey-Engley Neutralizing Broth M 1062
Appearance of the products:	white tablets
Product diluent:	hard water
Test concentration:	1 tablet/ 1.5 l (3.33 g/1.5 l = 2.22 g/l, 0.222%)
Test concentration:	1 tablet/ 3 l (3.33 g/3 l = 1.11 g/l, 0.111%)
Contact time:	15 min 30 min
Interfering substances:	0.3 g/l BSA (clean conditions)
Test organisms:	*Mycobacterium terrae* My 238/80 CNCTC, ATCC „15755" *Mycobacterium avium* ATCC 15769

Test procedure:

1. Preparation of test suspension
2. Counting of test suspension
3. Quantitative suspension test

Note:
Mycobactericidal activity – the capability of a product to produce a reduction in the number of viable cells of *Mycobacterium terrae* and *Mycobacterium avium* under defined conditions by at least 4 orders (10^4).
Tuberculocidal activity - the capability of a product to produce a reduction in the number of viable cells of *Mycobacterium terrae* under defined conditions by at least 4 orders (10^4).
$\Delta \log N = \log N_{\text{cfu/ml test suspension}} - \log N_{\text{cfu/ml after test procedure}}$

The standard:
ČSN EN 14348 Chemical disinfectants and antiseptic – Quantitative suspension test for the evaluation of mycobactericidal activity of chemical disinfectants in the medical area including instrument disinfectants - Test method and requirements (phase 2/step 1)

Description: *Testing the efficacy of chemical disinfectants and antiseptics* – SOP-M-19-00

Sample ID: 1645/2006
Rep No: 48
Sample name: **CHLORAMIX DT**
Sampled: by client
Sampling point: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín
Client: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín

Sampling date: 11.8.2006
Sample delivered: 11.8.2006
Testing date: 22.8.2006-8.11.2006
Delivered amount: 500 g
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Tab No. 23 Testing the efficacy of chemical disinfectant **CHLORAMIX DT** on *Mycobacterium avium*

Test concentration (g/l) / contact time (min) / conditions	Dilution after test procedure	Count CFU/ml	N	log N	Δlog N
2.22/15/clean	10^{-2}	8	$8.0 \cdot 10^{2}$	2.90	6.42
1.11/30/clean	10^{-3}	146	$1.46 \cdot 10^{5}$	5.16	4.16
Test suspension	10^{-8}	21	$2.1 \cdot 10^{9}$	9.32	

Tab No. 24 Testing the efficacy of chemical disinfectant **CHLORAMIX DT** on *Mycobacterium terrae*

Test concentration (g/l) / contact time (min) / conditions	Dilution after test procedure	Count CFU/ml	N	log N	Δlog N
2.22/15/clean	10^{-4}	18	$1.8 \cdot 10^{5}$	5.26	5.19
1.11/30/clean	10^{-5}	32	$3.2 \cdot 10^{6}$	6.51	3.94
Test suspension	10^{-9}	28	$2.8 \cdot 10^{10}$	10.45	

$\Delta\log N = \log N_{\text{cfu/ml test suspension}} - \log N_{\text{cfu/ml after test procedure}}$

Tab No. 25 The efficacy of chemical disinfectant **CHLORAMIX DT** on test strains – mycobactericidal and tuberculocidal activity

Mycobactericidal and tuberculocidal activity of the product (ČSN EN 14348)						
Strain	Test temperature [°C]	Contact time [min]	Product test concentrations [g/l (%)]	Interfering substances - conditions	Δlog N ČSN EN 14348	Δlog N
Mycobacterium avium	20	15	2.22 (0.222)	clean	> 4	> 4
Mycobacterium terrae	20	15	2.22 (0.222)	clean	> 4	> 4
Mycobacterium avium	20	30	1.11 (0.111)	clean	> 4	> 4
Mycobacterium terrae	20	30	1.11 (0.111)	clean	> 4	< 4

Prepared by: Hana Konevalíková, Lab Assistant

Description: *Testing the efficacy of chemical disinfectants and antiseptics* – SOP-M-19-00

Sample ID: 1645/2006
Rep No: 48
Sample name: **CHLORAMIX DT**
Sampled: by client
Sampling point: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín
Client: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín

Sampling date: 11.8.2006
Sample delivered: 11.8.2006
Testing date: 22.8.2006-8.11.2006
Delivered amount: 500 g
Batch No: 06-018-0-LO-00:29
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Interpretation:
Results of tests are in Tabs No. 1-25
According to ČSN EN 13727 and ČSN EN 1276 the tested product **CHLORAMIX DT,** batch No. 06-018-0-LO-00:29, in the concentration 1 tablet/5 l (3.33 g/5 l = 0.66 g/l, 0.066%) and contact time 15 min, diluted in hard water, under dirty conditions, at temperature 20 °C ± 1 °C, **proved** by the method of neutralizying dilutation to decrease the number of alive microbes *Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus, Enterococcus hirae* by 5 (lg) orders.

According to ČSN EN 13624 and ČSN EN 1650 the tested product **CHLORAMIX DT,** batch No. 06-018-0-LO-00:29, in the concentration 1 tablet/5 l (3.33 g/5 l = 0.66 g/l, 0.066%) and contact time 15 min, and in the concentration 1 tablet/3 l (3.33 g/3 l = 1.11 g/l, 0.111%) and contact time 15 min, diluted in hard water, under dirty conditions, at temperature 20 °C ± 1 °C, **proved** by the method of neutralizying dilutation to decrease the number of alive microbes *Candida albicans, Aspergillus niger* by 4 (lg) orders.

According to ČSN EN 13697 the tested product **CHLORAMIX DT,** batch No. 06-018-0-LO-00:29, in the concentration 1 tablet/5 l (3.33 g/5 l = 0.66 g/l, 0.066%) and contact time 15 min, diluted in hard water, under dirty conditions, and in the concentration 1 tablet/10 l (3.33 g/10 l = 0.33 g/l, 0.033%) and contact time 15 min diluted in hard water, under clean conditions, at temperature 20 °C ± 1 °C, **proved** by the method of neutralizying dilutation on carriers to decrease the number of alive microbes *Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus, Enterococcus hirae* by 4 (lg) orders.

According to ČSN EN 13697 the tested product **CHLORAMIX DT,** batch No. 06-018-0-LO-00:29, in the concentration 1 tablet/5 l (3.33 g/5 l = 0.66 g/l, 0.066%) and contact time 15 min, diluted in hard water, under dirty conditions, and in the concentration 1 tablet/10 l (3.33 g/10 l = 0.33 g/l, 0.033%) and contact time 15 min diluted in hard water, under clean conditions, at temperature 20 °C ± 1 °C, **proved** by the method of neutralizying dilutation on carriers to decrease the number of alive microbes *Candida albicans, Aspergillus niger* by 3 (lg) orders.

According to ČSN EN 14476 the tested product **CHLORAMIX DT,** batch No. 06-018-0-LO-00:29, in the concentration 1 tablet/1.5 l (3.33 g/1.5 l = 2.22 g/l, 0.222%) and contact time 15 min, diluted in hard water, under dirty conditions, and in the concentration 1 tablet/5 l (3.33 g/5 l = 0.66 g/l, 0.066%) and contact time 15 min, diluted in hard water, under clean conditions, at temperature 20 °C ± 1 °C, **proved** by the method of virus titration on monolayers of cells in cell culture tubes to reduce in the number of infectious virus particles (*poliovirus* type 1, LSc-2ab) under defined conditions by at least 4 orders (10^4).

According to ČSN EN 14476 the tested product **CHLORAMIX DT,** batch No. 06-018-0-LO-00:29, in the concentration 1 tablet/5 l (3.33 g/5 l = 0.66 g/l, 0.066%) and contact time 15 min, diluted in hard water, under clean conditions, at temperature 20 °C ± 1 °C, **proved** by the method of virus titration on monolayers of cells in cell culture tubes to reduce in the number of infectious virus particles (*adenovirus* type 5) under defined conditions by at least 4 orders (10^4).

According to SOP-M-19-00-H the tested product **CHLORAMIX DT,** batch No. 06-018-0-LO-00:29, in the concentration 1 tablet/1.5 l (3.33 g/1.5 l = 2.22 g/l, 0.222%) and contact time 15 min, diluted in hard water, under dirty conditions, at temperature 20 °C ± 1 °C, **proved** by the method of virus titration on monolayers of cells in cell culture tubes to reduce in the number of infectious virus particles (BVDV strain NADL ATCC-VR-534) under defined conditions by at least 4 orders (10^4).

Description: *Testing the efficacy of chemical disinfectants and antiseptics* – SOP-M-19-00

Sample ID: 1645/2006
Rep No: 48
Sample name: **CHLORAMIX DT**
Sampled: by client
Sampling point: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín
Client: BOCHEMIE, s.r.o., Lidická 326, 735 95 Bohumín

Sampling date: 11.8.2006
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Batch No: 06-018-0-LO-00:29
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According to ČSN EN 14204 the tested product **CHLORAMIX DT,** batch No. 06-018-0-LO-00:29, in the concentration 1 tablet/1.5 l (3.33 g/1.5 l = 2.22 g/l, 0.222%) and contact time 15 min, diluted in hard water, under clean conditions, at temperature 20 °C ± 1 °C, **proved** by the method of neutralizying dilutation to decrease the number of alive microbes *Mycobacterium terrae* and *Mycobacterium avium* by 4 (lg) orders.

According to ČSN EN 14348 the tested product **CHLORAMIX DT,** batch No. 06-018-0-LO-00:29, in the concentration 1 tablet/3 l (3.33 g/3 l = 1.11 g/l, 0.111%) and contact time 30 min, diluted in hard water, under clean conditions, at temperature 20 °C ± 1 °C, **proved** by the method of neutralizying dilutation to decrease the number of alive microbes *Mycobacterium terrae* by 4 (lg) orders.

Conclusion:
The product **CHLORAMIX DT** is capable of reducing the number of viable bacterial cells, the number of viable vegetative yeast cells and mould spores, the number of viable mycobacterial cells relevant organisms under defined conditions to the declared values and consequently may be called bactericidal, fungicidal, mycobactericidal and tuberculocidal.
The product **CHLORAMIX DT** is capable of reducing the number of infectious *poliovirus* and *adenovirus* particles under defined conditions to the declared values and consequently may be called virucidal.
The product **CHLORAMIX DT** is capable of reducing the number of infectious BVDV strain NADL ATCC-VR-534 particles under defined conditions to the declared values and consequently may be called virucidal on BVDV.

4.2.2008, Hodonín

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Ing. Jana Šlitrová, the leader of study