

Copy No.: 1  
Issue No.: 2

Test report No. D143/2013

DETERMINATION OF BACTERICIDAL (EN 13697) AND YEASTICIDAL (EN 13697) ACTIVITY OF THE PRODUCT **QUATRODES FORTE**

Sample ID: D143/2013

Sample name: **Quatrodes Forte**

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

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From pages: 7

Incoming date:  
2.9.2013

Delivery date:  
20.2.2014

Hodonín, 20.2.2014



Zuzana Matuskova, Head of Laboratory

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

Sample ID: D143/2013  
Rep No: 143  
Sample name: **Quatrodes Forte**  
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

Sampling date: 29.8.2013  
Sample delivered: 2.9.2013  
Testing date: 13.9. – 7.10.2013  
Delivered amount: 400 ml  
Batch No: A-28-SIE-15  
Page: 2

Subject of testing:

Determination of bactericidal and yeasticidal activity of the product.

Identification of the sample:

Name of the product:	<b>Quatrodes Forte</b>
Batch number:	A-28-SIE-15
Date of manufacture:	28.08.2013
Expiry date:	02.2016
Manufacturer:	Medi-Sept Sp. z o.o., Konopnica 159c, 210 30 Motycz, Poland
Incoming date:	2.9.2013
Storage conditions:	stated by the manufacturer
Active ingredients:	100 g contains:
	3,76 g N-(3-Aminopropyl)-N-dodecylpropane-1,3-diamine (CAS: 2372-82-9)
	3,39 g N,N-Didecyl-N-methyl-poly(oxyethyl)ammonium propionate (CAS: 94667-33-1)

Experimental conditions:

	<b>Quantitative test on carriers for the evaluation of bactericidal activity SOP-M-22-12 (EN 13697)</b>
Period of analysis:	24.9. – 25.9.2013
Test temperature:	18 °C ± 1 °C až 25 °C ± 1 °C
Test method:	dilution neutralization method
Neutralization medium:	Dey-Engley Neutralizing Broth M 1062
Product diluent:	hard water
Appearance of the products:	yellow liquid
Test concentration:	1%
Contact time:	15 min
Interfering substances:	0.3 g/l BSA (clean conditions)
Test organisms:	<i>Escherichia coli</i> ATCC 10536 <i>Pseudomonas aeruginosa</i> ATCC 15442 <i>Staphylococcus aureus</i> ATCC 6538 <i>Enterococcus hirae</i> ATCC 10541
Incubation conditions:	37 °C ± 1 °C, 24 hours

Test procedure:

1. Preparation of the test suspension
2. Preparation of product test solutions
3. Quantitative carrier test
4. Incubation and calculation
5. Expression and interpretation of results

Note:

Bactericidal activity – the capability of a product to produce a reduction in the number of viable bacterial cells of relevant organisms on carriers under defined conditions by at least 4 orders ( $10^4$ ). The drying time: 30-35 min

The standard:

EN 13697 Chemical disinfectants and antiseptics – Quantitative non-porous surface test for the evaluation of bactericidal and/or fungicidal activity of chemical disinfectants used in food, industrial, domestic and institutional areas - Test method and requirements without mechanical action (phase 2/step 2) August 2001

Description: Testing the efficacy of chemical disinfectants and antiseptics

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

1. Testing the efficacy of chemical disinfectant **Quatrodes Forte** on carriers – bactericidal activity

Tab No. 1.1 Verification of methodology, clean conditions

Test organisms	Test suspension N	Validation test	
		NT Neutralization test	NC Neutralization control
<i>Escherichia coli</i> ATCC 10536	10 <sup>-6</sup> : >300, >300 10 <sup>-7</sup> : 45, 46 N : 7.36	10 <sup>-3</sup> : >300, >300 10 <sup>-4</sup> : 173, 231 10 <sup>-5</sup> : 16, 21 NT : 7.31	10 <sup>-3</sup> : >300, >300 10 <sup>-4</sup> : 170, 226 10 <sup>-5</sup> : 17, 19 NC : 7.30
<i>Pseudomonas aeruginosa</i> ATCC 15442	10 <sup>-6</sup> : >300, >300 10 <sup>-7</sup> : 48, 52 N : 7.40	10 <sup>-3</sup> : >300, >300 10 <sup>-4</sup> : 229, 244 10 <sup>-5</sup> : 21, 25 NT : 7.37	10 <sup>-3</sup> : >300, >300 10 <sup>-4</sup> : 237, 245 10 <sup>-5</sup> : 22, 26 NC : 7.38
<i>Staphylococcus aureus</i> ATCC 6538	10 <sup>-6</sup> : >300, >300 10 <sup>-7</sup> : 41, 49 N : 7.35	10 <sup>-3</sup> : >300, >300 10 <sup>-4</sup> : 201, 233 10 <sup>-5</sup> : 20, 22 NT : 7.34	10 <sup>-3</sup> : >300, >300 10 <sup>-4</sup> : 216, 243 10 <sup>-5</sup> : 21, 23 NC : 7.36
<i>Enterococcus hirae</i> ATCC 10541	10 <sup>-6</sup> : >300, >300 10 <sup>-7</sup> : 45, 49 N : 7.37	10 <sup>-3</sup> : >300, >300 10 <sup>-4</sup> : 209, 237 10 <sup>-5</sup> : 22, 25 NT : 7.35	10 <sup>-3</sup> : >300, >300 10 <sup>-4</sup> : 172, 228 10 <sup>-5</sup> : 19, 23 NC : 7.30

$$N = \log_{10} [\{0.05 \cdot (x + x')\} / 2 \cdot d]$$

where x and x' are paired values for which the mean of the value falls between 50 and 300 colonies, d is the dilution factor for the dilution taken into account

$$NC \text{ or } NT = \log_{10} [\{10 \cdot (y + y')\} / 2 \cdot d]$$

where y and y' are paired values for which the mean of the value falls between 50 and 300 colonies, d is the dilution factor for the dilution taken into account

Tab No. 1.2 Testing the efficacy of chemical disinfectant **Quatrodes Forte** on test strain, clean conditions

Test organisms	Water control Nc	Test procedure Nd at concentrations (%) / contact time (min)
		1/15
<i>Escherichia coli</i> ATCC 10536	10 <sup>-3</sup> : >300, >300 10 <sup>-4</sup> : 175, 224 10 <sup>-5</sup> : 19, 24 10 <sup>-6</sup> : 2, 3 Nc : 7.30 Nts : 207	10 <sup>0</sup> : 0, 0 10 <sup>-1</sup> : 0, 0 10 <sup>-2</sup> : 0, 0 Nd : < 0.10 Nts : 0 <b>ME : ≥ 7.20</b>
<i>Pseudomonas aeruginosa</i> ATCC 15442	10 <sup>-3</sup> : >300, >300 10 <sup>-4</sup> : 245, 249 10 <sup>-5</sup> : 22, 26 10 <sup>-6</sup> : 2, 3 Nc : 7.39 Nts : >300	10 <sup>0</sup> : 0, 0 10 <sup>-1</sup> : 0, 0 10 <sup>-2</sup> : 0, 0 Nd : < 0.10 Nts : 0 <b>ME : ≥ 7.29</b>
<i>Staphylococcus aureus</i> ATCC 6538	10 <sup>-3</sup> : >300, >300 10 <sup>-4</sup> : 236, 240 10 <sup>-5</sup> : 21, 25 10 <sup>-6</sup> : 2, 2 Nc : 7.38 Nts : >300	10 <sup>0</sup> : 0, 0 10 <sup>-1</sup> : 0, 0 10 <sup>-2</sup> : 0, 0 Nd : < 0.10 Nts : 0 <b>ME : ≥ 7.28</b>
<i>Enterococcus hirae</i> ATCC 10541	10 <sup>-3</sup> : >300, >300 10 <sup>-4</sup> : 240, 244 10 <sup>-5</sup> : 23, 25 10 <sup>-6</sup> : 2, 3 Nc : 7.38 Nts : >300	10 <sup>0</sup> : 0, 0 10 <sup>-1</sup> : 0, 0 10 <sup>-2</sup> : 0, 0 Nd : < 0.10 Nts : 0 <b>ME : ≥ 7.28</b>

$$Nc \text{ or } Nd = \log_{10} [\{10 \cdot (a + a')\} / 2 \cdot d]$$

where a and a' are paired values for which the mean of the value falls between 50 and 300 colonies, d is the dilution factor for the dilution taken into account

$$\text{Microbicidal effect } ME = Nc - Nd$$



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

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Sampled: by client  
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Experimental conditions:

Period of analysis:

Test temperature:

Test method:

Neutralization medium:

Product diluent:

Appearance of the products:

Test concentration:

Contact time:

Interfering substances:

Test organisms:

Incubation conditions:

**Quantitative test on carriers for the evaluation of fungicidal activity SOP-M-22-12 (EN 13697)**

13.9. – 16.9.2013 *C. albicans*

3.10. – 7.10.2013 *A. brasiliensis (niger)*

18 °C ± 1 °C až 25 °C ± 1 °C

dilution neutralization method

Dey-Engley Neutralizing Broth M 1062

hard water

yellow liquid

1%

15 min

0.3 g/l BSA (clean conditions)

*Candida albicans*

ATCC 10231

30 °C ± 1 °C, 48 hours and additional period of 24 or 48 hours

Test procedure:

1. Preparation of the test suspension
2. Preparation of product test solutions
3. Quantitative carrier test
4. Incubation and calculation
5. Expression and interpretation of results

Note:

Fungicidal activity – the capability of a product to produce a reduction in the number of viable fungi of relevant organisms on carriers under defined conditions by at least 3 orders ( $10^3$ ).

Yeasticidal activity – the capability of a product to produce a reduction in the number of viable yeast cells of relevant test organisms under defined conditions by at least 3 orders ( $10^3$ ).

The standard:

EN 13697 Chemical disinfectants and antiseptics – Quantitative non-porous surface test for the evaluation of bactericidal and/or fungicidal activity of chemical disinfectants used in food, industrial, domestic and institutional areas - Test method and requirements without mechanical action (phase 2/step 2) August 2001

Description: Testing the efficacy of chemical disinfectants and antiseptics

Sample ID: D143/2013  
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2. Testing the efficacy of chemical disinfectant **Quatroles Forte** on carriers – fungicidal activity

Tab No. 2.1 Verification of methodology, clean conditions

Test organisms	Test suspension N	Validation test	
		NT Neutralization test	NC Neutralization control
<i>Candida albicans</i> ATCC 10231	10 <sup>-5</sup> : >300, >300 10 <sup>-6</sup> : 48, 50 N : 6.39	10 <sup>-3</sup> : 225, 243 10 <sup>-4</sup> : 21, 24 10 <sup>-5</sup> : 2, 3 NT : 6.37	10 <sup>-3</sup> : 218, 230 10 <sup>-4</sup> : 19, 22 10 <sup>-5</sup> : 1, 3 NC : 6.35

$N = \log_{10} [\{0.05 \cdot (x + x')\} / 2 \cdot d]$  where x and x' are paired values for which the mean of the value falls between 50 and 300 colonies, d is the dilution factor for the dilution taken into account

NC or NT =  $\log_{10} [\{10 \cdot (y + y')\} / 2 \cdot d]$  where y and y' are paired values for which the mean of the value falls between 50 and 300 colonies, d is the dilution factor for the dilution taken into account

Tab No. 2.2 Testing the efficacy of chemical disinfectant **Quatroles Forte** on test strain, clean conditions

Test organisms	Water control Nc	Test procedure Nd at concentrations (%) / contact time (min)
		1/15
<i>Candida albicans</i> ATCC 10231	10 <sup>-2</sup> : >300, >300 10 <sup>-3</sup> : 217, 241 10 <sup>-4</sup> : 21, 25 10 <sup>-5</sup> : 2, 2 Nc : 6.36 Nts : >300	10 <sup>0</sup> : 0, 0 10 <sup>-1</sup> : 0, 0 10 <sup>-2</sup> : 0, 0 Nd : < 0.10 Nts : 0 <b>ME : ≥ 6.26</b>

$Nc$  or  $Nd = \log_{10} [\{10 \cdot (a + a')\} / 2 \cdot d]$  where a and a' are paired values for which the mean of the value falls between 50 and 300 colonies, d is the dilution factor for the dilution taken into account

Microbicidal effect ME = Nc – Nd

Description: Testing the efficacy of chemical disinfectants and antiseptics

Sample ID: D143/2013

Rep No: 143

Sample name: **Quatroles Forte**

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Batch No: A-28-SIE-15

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### 3. Evaluation of bactericidal and yeasticidal activity of the product **Quatroles Forte** on carriers

Tab No. 3.1 The efficacy of chemical disinfectant **Quatroles Forte** on test strains – bactericidal and yeasticidal activity on carriers

Bactericidal and yeasticidal activity of the product on carriers (EN 13697)						
Strain	Test temperature [°C]	Contact time [min]	Product test concentrations [%]	Interfering substances - conditions	ME EN 13697	ME
<i>Escherichia coli</i> ATCC 10536	18-25	15	1	clean	≥ 4	> 4
<i>Pseudomonas aeruginosa</i> ATCC 15442	18-25	15	1	clean	≥ 4	> 4
<i>Staphylococcus aureus</i> ATCC 6538	18-25	15	1	clean	≥ 4	> 4
<i>Enterococcus hirae</i> ATCC 10541	18-25	15	1	clean	≥ 4	> 4
<i>Candida albicans</i> ATCC 10231	18-25	15	1	clean	≥ 3	> 3

Microbicidal effect ME = Nc – Nd

Prepared by: Hana Konevalíková, Lab Technician

Description: Testing the efficacy of chemical disinfectants and antiseptics

Sample ID: D143/2013

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Sample name: **Quatrodes Forte**

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Interpretation:

Results of tests are in Tabs.

The tested product **Quatrodes Forte**, batch No. A-28-SIE-15, in the concentration 1%, diluted in hard water, and the contact time 15 min under clean conditions at temperature  $18\text{ }^{\circ}\text{C} \pm 1\text{ }^{\circ}\text{C}$  to  $25\text{ }^{\circ}\text{C} \pm 1\text{ }^{\circ}\text{C}$  by the dilution neutralization method **decreased** on carriers the number of alive microbes *Escherichia coli* ATCC 10536, *Pseudomonas aeruginosa* ATCC 15442, *Staphylococcus aureus* ATCC 6538, *Enterococcus hirae* ATCC 10541 by at least 4 (lg) orders (EN 13697).

According to EN 13697 the tested product **Quatrodes Forte**, batch No. A-28-SIE-15, in the concentration 1%, diluted in hard water, and the contact time 15 min under clean conditions at temperature  $18\text{ }^{\circ}\text{C} \pm 1\text{ }^{\circ}\text{C}$  to  $25\text{ }^{\circ}\text{C} \pm 1\text{ }^{\circ}\text{C}$  by the dilution neutralization method, **decreased** on carriers the number of alive microbes *Candida albicans* ATCC 10231 by at least 3 (lg) orders.

Conclusion:

The product **Quatrodes Forte** is capable of reducing the number of viable bacterial and vegetative yeast cells of the relevant organisms on carriers under defined conditions to the declared values, and consequently, may be called bactericidal and yeasticidal on carriers.

20.2.2014, Hodonín

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Ing. Jana Štířová, Leader of Study  
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