



**SILLIKER report for ENVIROPRODUCTS**  
**Validation of the efficiency of Microbial removal process**  
**and Evaluation of bacteria transfer when using E-Cloths**  
**products and water**

**Study performed on: May and August 2010**

<b>By</b>	
<b>SILLIKER S.A.S</b> Laboratoire de Microbiologie - Unité R&D 10, rue les Châteaux Saint Sylvère 95011 CERGY Cedex - FRANCE	<b>Aurélie PERNOT</b> <i>Research Engineer</i> Telephone : 33 (0)1 30 75 61 56 E-mail : <a href="mailto:aurelie.pernot@silliker.fr">aurelie.pernot@silliker.fr</a>
<b>For</b>	
<b>EnviroProducts</b> Ltd, East Barn, Furnace Farm Furnace Lane, Lamberhurst Kent. TN3 8LE	<b>Laurence SMITH</b> <i>Commercial Director</i> Phone: 01892 893130 E-mail : <a href="mailto:laurence.smith@e-cloth.com">laurence.smith@e-cloth.com</a>

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## I. SUMMARY

This report shows the results and the technical protocol of the SILLIKER studies performed for ENVIROPRODUCTS.

The microbial validation of E-Cloths study consists in performing trials in order to evaluate the efficiency of the E-Cloths to remove micro-organisms by using only E-Cloths micro-fibre and water. A laminate surface is contaminated by microorganisms, and then wiped by E-cloth micro-fibre using water or other competitor products (using chemicals). The contamination on the surface is checked before (initial concentration) and after (residual concentration) cleaning with E-Cloth micro-fibre or competitors products. For each test controls are performed. The results obtained with E-Cloths are shown in percentage of microbial removal and compared to those obtained with competitor cleaning methods.

The study of the bacteria transfer evaluation consists in cleaning a contaminated surface with E-Cloth, wash E-Cloth with water and wipe a sterile surface with it, in order to evaluate the number of bacteria left by the cloth on a sterile surface.

## II. MATERIALS AND METHODS FOR MICROBIAL VALIDATION OF E-CLOTHS

### II.1. PROTOCOL GENERAL

In order to mimic realistic conditions of E-Cloths cleaning with water, 100 cm<sup>2</sup> surfaces are prepared in the laboratory. Then, surfaces are contaminated by one of the micro-organisms of the study. Microbial concentration is checked in this surface with wipes.

Initial concentration is determined by a wiping after the first spreading of the inoculum on the surface. Then, the residual concentration on the surface is determined by a taking after wiping with the E-Cloths or competitor's product.

E-Cloths use only water while competitors use chemicals or other non chemical products.

After each spreading of inoculum, the surface is left for necessary time to get dry the inoculum. The difference between the initial contamination and the residual contamination is correlated to percentage of microbial removal.

## II.2. PRODUCTS

### II.2.1. TWO BACTERIA

<i>Protocols</i>	<i>Additives</i>
Micro Fibre E-cloth General Purpose using water	Water
Conventional Tesco dish cloth 4 for £1-50	Flash All Purpose Lemon All for one (yellow)
Conventional J-cloth Tesco own label	Cif power cream kitchen
Tesco dish cloth 4 for £1-50	Ecover all Purpose Cleaner

### II.2.2. THREE BACTERIA

<i>Additives</i>	<i>Protocols</i>
<b>Water</b>	Micro Fibre E-cloth Glass & Polishing
<b>Chemicals</b>	Tesco Cotton dish cloth 4 for £1-50 cloth using Cif Kitchen Spray
	Conventional J-cloth using Flash All purpose lemon
	USA Paper Towel Pledge Bounty using Cif Kitchen Spray Power Cream
	Dettol Wipes (UK)
	E-cloth General Purpose using Lysol
<b>Non chemical additives</b>	Micro Fibre Scotchbrite Kitchen Cloth using Ecover all Purpose Cleaner
	Tesco dish cloth 4 for £1-50 using Tesco Natural Kitchen Liquid Cleaner £1-50 500ml
	Conventional J-cloth Tesco Own Brand using Ecover all Purpose Cleaner
<b>Disinfectant</b>	Conventional Cotton cloth Tesco dish cloth 4 for £1-50 using Dettol
	Conventional Cotton cloth using Lysol (US)

### Test mops

<b>Additives</b>	<b>TEST Mops</b>
<b>Water</b>	Micro Fibre E-cloth Damp Head using water
<b>Flash floor All in One (Blue)</b>	Conventional Rag mop using Flash floor All in One (Blue)
<b>Method Liquid</b>	Micro Fibre Method using Method Liquid

### II.3. SURFACE

- Laminate 100cm<sup>2</sup> (10cmx10cm)

### II.4. CHOICE OF STRAINS

Selection of the strains is carried out according to the attempted use of the E-Cloths. The table below shows the list of organisms for artificial contaminations.

Organisms	Strains
<b>Hygienic indicator Bacteria</b>	<i>Escherichia coli</i>
<b>Safety Bacteria</b>	<i>Listeria monocytogenes</i>
<b>Mould</b>	<i>Aspergillus niger</i>

**Table 1 : Strains used to artificial contamination of surfaces**

### II.5. PREPARATION OF THE STRAINS

Two steps are required in the preparation of the strains:

1. First subculture: is prepared in a medium and at temperature favourable to optimal growth of each inoculated organism and sufficient time to reach the beginning of the stationary phase.
2. Second subculture: prepared and incubated at temperature close to the room temperature (20 °C)

Each strain will be used separately.

### II.6. CONTAMINATION OF THE SURFACES

In order to get harmonised results, a surface of 100 cm<sup>2</sup> is artificially contaminated. The inoculation of micro-organisms in the surface is realised by spreading the same volume on the test surface (1 mL). 3 separated surfaces are used for each test. When the inoculum is spread, one surface is used to check initial contamination (A). The second surface is cleaned by E-cloths then used to determine residual contamination (B). The third surface is not cleaned and used as control (C).

The initial concentration targeted by the artificial contamination is about 10<sup>6</sup> organisms in the 100 cm<sup>2</sup> area.

### II.7. MICROBIOLOGICAL METHODS

Counting of the microbiological takings is realised with ISO standard methods. Details of these methods are given in the table below:

Organism	Microbiological methods
<i>Escherichia coli</i>	NF ISO 16649-2
<i>Listeria monocytogenes</i>	EN ISO 11290-2
<i>Aspergillus niger</i>	NF V08-059

**Table 2 : Microbiological methods**

## II.8. PROTOCOL

### II.8.1. ARTIFICIAL CONTAMINATION OF THE SURFACES

Artificial contamination of surfaces is done by spreading a volume of the inoculum on the surface. The surface is left for about 30 minutes to dry.

The concentration targeted in the contaminated surfaces is about  $10^6$  cfu/100cm<sup>2</sup>.

### II.8.2. CLEANING OF THE SURFACES

E-Cloth and competitor products are used as specified by EnviroProducts.

### II.8.3. MICROBIOLOGICAL TAKINGS

After cleaning, the surface is wiped by sterile wipes used to microbial takings that are used in the laboratory.

## III. RESULTS FOR MICROBIAL VALIDATION OF E-CLOTHS

### III.1. ESCHERICHIA COLI

#### III.1.1. TEST WITH CLOTHS

Protocols	Additives	Result (cfu/100 cm <sup>2</sup> )	% of microbial removal
Control 1	-	7 000 000	-
Control 2	-	6 200 000	-
Control 3	-	6 100 000	-
Control 4	-	<100	-
Control 5	-	<100	-
Control 6	-	~1 300 000	-
Micro Fibre <b>E-cloth</b> General Purpose	Water	4000	<b>99.9355</b>
Conventional Tesco dish cloth 4 for £1-50	Flash All Purpose Lemon All for one (yellow)	1900	99.9694
Conventional J-cloth Tesco own label	Cif power cream kitchen	<10	99.9998
Tesco dish cloth 4 for £1-50	Ecover all Purpose Cleaner	360	99.9942

Additives	Protocols	Additives	Result (cfu/100cm <sup>2</sup> )	% of microbial removal
<b>Water</b>	Micro Fibre <b>E-cloth</b> Glass & Polishing	Water	<10	<b>99.9998</b>
<b>Chemicals</b>	Tesco Cotton dish cloth 4 for £1-50 cloth	Cif Kitchen Spray	<10	99,9992
	Conventional J-cloth	Flash All purpose lemon	~20	99,9985
	USA Paper Towel Pledge Bounty	Cif Kitchen Spray Power Cream	<10	99,9992
	Dettol Wipes (UK)	-	~30	99,9977
	<b>E-cloth</b> General Purpose	Lysol	<10	<b>99,9992</b>
<b>Non chemical additives</b>	Micro Fibre Scotchbrite Kitchen Cloth	Ecover all Purpose Cleaner	<10	99,9992
	Tesco dish cloth 4 for £1-50	Tesco Natural Kitchen Liquid Cleaner £1-50 500ml	<10	99,9992
	Conventional J-cloth Tesco Own Brand	Ecover all Purpose Cleaner	~60	99,9954
<b>Disinfectant</b>	Conventional Cotton cloth Tesco dish cloth 4 for £1-50	Dettol	<10	99,9992
	Conventional Cotton cloth	Lysol (US)	<10	99,9992

### III.1.2. TEST MOPS

TEST Mops	Additives	Result (cfu/100cm <sup>2</sup> )	% of microbial removal
Micro Fibre <b>E-cloth</b> Damp Head	Water	2500	<b>99,8077</b>
Conventional Rag mop	Flash floor All in One (Blue)	39 000	97,0000
Micro Fibre Method	Method Liquid	490	99,9623

Median of the controls (day 1: control 1,2 and 3) = 6 200 000 cfu/100cm<sup>2</sup>

Median of the controls (day 2: control 4,5 and 6) = ~1 300 000 cfu/100cm<sup>2</sup>

Tests performed day 1 are those in the first table and in the second table, the test with water (without sample 1, 2 and 3). Tests performed day 2 are the tests in the second table from the test "sample 1", and test mops.

Percentage of microbial removal = ((Median – residual concentration) / median) x 100

#### Interpretation:

In term *Escherichia coli* removal, the results of the study show that efficiency of cleaning with E-Cloth micro-fibre and just water is close to the efficiency of competitor cloths using disinfectant.



## III.2. LISTERIA MONOCYTOGENES

### III.2.1. TEST WITH CLOTHS

Protocols	Additives	Result (cfu/100cm <sup>2</sup> )	% of microbial removal
Control 1	-	9 600 000	-
Control 2	-	~12 000 000	-
Control 3	-	8 900 000	-
Control 4	-	5 000 000	-
Control 5	-	4 500 000	-
Control 6	-	5 600 000	-
Control 7	-	250 000	-
Control 8	-	170 000	-
Control 9	-	180 000	-
Micro Fibre <b>E-cloth</b> General Purpose	Water	92 000	<b>99,0417</b>
Conventional Tesco dish cloth 4 for £1-50	Flash All Purpose Lemon All for one (yellow)	15 000	99,8438
Conventional J-cloth Tesco own label	Cif power cream kitchen	38 000	99,6042
Tesco dish cloth 4 for £1-50	Ecover all Purpose Cleaner	5 200	99,9458

Additives	Protocols	Additives	Result (cfu/100cm <sup>2</sup> )	% of microbial removal
<b>Water</b>	Micro Fibre <b>E-cloth</b> Glass & Polishing	Water	4 800	<b>99,9500</b>
<b>Chemicals</b>	Tesco Cotton dish cloth 4 for £1-50 cloth	Cif Kitchen Spray	35 000	99,3000
	Conventional J-cloth	Flash All purpose lemon	22 000	99,5600
	USA Paper Towel Pledge Bounty	Cif Kitchen Spray Power Cream	3 500	99,9300
	Dettol Wipes (UK)	-	~400	99,9920
	<b>E-cloth</b> General Purpose	Lysol	<100	<b>99,9980</b>
<b>Non chemical additives</b>	Micro Fibre Scotchbrite Kitchen Cloth	Ecover all Purpose Cleaner	<100	99,9980
	Tesco dish cloth 4 for £1-50	Tesco Natural Kitchen Liquid Cleaner £1-50 500ml	~400	99,9920
	Conventional J-cloth Tesco Own Brand	Ecover all Purpose Cleaner	~100	99,9980
<b>Disinfectant</b>	Conventional Cotton cloth Tesco dish cloth 4 for £1-50	Dettol	<100	99,9980
	Conventional Cotton cloth	Lysol (US)	<100	99,9980

### III.2.2. TEST MOPS

TEST Mops	Additives	Result (cfu/100cm <sup>2</sup> )	% of microbial removal
Micro Fibre <b>E-cloth</b> Damp Head	Water	97 000	<b>98,0600</b>
Conventional Rag mop	Flash floor All in One (Blue)	68 000	98,6400
Micro Fibre Method	Method Liquid	52 000	98,9600

Median of the controls (day 1: control 1,2 and 3) = 9 600 000 cfu/100cm<sup>2</sup>

Median of the controls (day 2: control 4,5 and 6) = 5 000 000 cfu/100cm<sup>2</sup>

Median of the controls (day 3: control 7,8 and 9) = 180 000 cfu/100cm<sup>2</sup>

Tests performed day 1 are in the first table and test with water in the second table (without sample 1, 2 and 3). Tests performed day 2 are those in the second table (chemical, non chemical additives and disinfectant) and test mops. The tests performed day 3 are the 3 analysis in the second table called "sample 1, sample 2, sample 3" using water.

Percentage of microbial removal = ((Median – residual concentration) / median) x 100

#### Interpretation:

In term of *Listeria monocytogenes* removal, the results of the study show that efficiency of cleaning with E-Cloth micro-fibre and just water is close to the efficiency of competitor cloths using chemicals and non chemicals additives. The *Listeria monocytogenes* removal is the best when disinfectant is used. For test mops, the results are close.

### III.3.ASPERGILLUS NIGER

#### III.3.1. TEST WITH CLOTHS

Protocols	Additives	Result (cfu/100cm <sup>2</sup> )	
Control 1	-	1600	
Control 2	-	~1400	
Control 3	-	1600	
Control 4	-	1800	
Control 5	-	1600	
Control 6	-	1500	
Micro Fibre <b>E-cloth</b> General Purpose	Water	92 000	99.3750
Conventional Tesco dish cloth 4 for £1-50	Flash All Purpose Lemon All for one (yellow)	15 000	99.3750
Conventional J-cloth Tesco own label	Cif power cream kitchen	38 000	99.3750
Tesco dish cloth 4 for £1-50	Ecover all Purpose Cleaner	5 200	99.3750

Median of the controls: = 1600 cfu/100cm<sup>2</sup>

All tests for *Aspergillus niger* were performed the same day.

Additives	Protocols	Additives	Result (cfu/100cm <sup>2</sup> )	% of microbial removal
<b>Water</b>	Micro Fibre <b>E-cloth</b> Glass & Polishing	Water	<10	<b>99.3750</b>
<b>Chemicals</b>	Tesco Cotton dish cloth 4 for £1-50 cloth	Cif Kitchen Spray	<10	99.3750
	Conventional J-cloth	Flash All purpose lemon	~10	99.3750
	USA Paper Towel Pledge Bounty	Cif Kitchen Spray Power Cream	<10	99.3750
	Dettol Wipes (UK)	-	<10	99.3750
	<b>E-cloth</b> General Purpose	Lysol	<10	<b>99.3750</b>
<b>Non chemical additives</b>	Micro Fibre Scotchbrite Kitchen Cloth	Ecover all Purpose Cleaner	~20	98.7500
	Tesco dish cloth 4 for £1-50	Tesco Natural Kitchen Liquid Cleaner £1-50 500ml	<10	99.3750
	Conventional J-cloth Tesco Own Brand	Ecover all Purpose Cleaner	<10	99.3750
<b>Disinfectant</b>	Conventional Cotton cloth Tesco dish cloth 4 for £1-50	Dettol	<10	99.3750
	Conventional Cotton cloth	Lysol (US)	~20	98.7500

### III.3.2. TEST MOPS

TEST Mops	Additives	Result (cfu/100cm <sup>2</sup> )	% of microbial removal
Micro Fibre <b>E-cloth</b> Damp Head	Water	<10	<b>99.3750</b>
Conventional Rag mop	Flash floor All in One (Blue)	<10	99.3750
Micro Fibre Method	Method Liquid	<10	99.3750

#### Interpretation:

In term of *Aspergillus niger* removal, the results of the study show that efficiency of cleaning with E-Cloth micro-fibre and just water is close to the efficiency of competitor cloths using chemicals and non chemicals additive, and disinfectants. For test mops, there is no difference between the efficacy of the different mops.

## IV. CONCLUSION

### Microbial validation of E-Cloths

In term *Escherichia coli* removal, the results of the study show that efficiency of cleaning with E-Cloth micro-fibre and just water is close to the efficiency of competitor cloths using disinfectant. According to the study, the best E-Cloth for *Escherichia coli* removal on laminate surface is Micro Fibre E-cloth Glass & Polishing using water.

In term of *Listeria monocytogenes* removal, the results of the study show that efficiency of cleaning with E-Cloth micro-fibre and just water is close to the efficiency of competitor cloths using chemicals and non chemicals additives.

In term of *Aspergillus niger* removal, the results of the study show that efficiency of cleaning with E-Cloth micro-fibre and just water is close to the efficiency of competitor cloths using chemicals and non chemicals additive, and disinfectants. According to the study, there is no difference between E-Cloth products for *Aspergillus niger* removal.

Test	% of <i>Escherichia coli</i> removal	% of <i>Listeria monocytogenes</i> removal	% of <i>Aspergillus niger</i> removal
Micro Fibre <b>E-cloth</b> General Purpose using Water	99.9355	99.0417	99.3750
Micro Fibre <b>E-cloth</b> Glass & Polishing using Water	99.9998	99.9500	99.3750
<b>E-cloth</b> General Purpose using Lysol	99.9992	99.9980	99.3750
Micro Fibre <b>E-cloth</b> Damp Head using Water	99.8077	98.0600	99.3750

## V. MATERIALS AND METHODS FOR EVALUATION OF BACTERIA TRANSFER

### V.1. BACTERIA

*Escherichia coli* is used to perform the test.

### V.2. SURFACE

A laminate surface is used. Contaminated and sterile surfaces are prepared.

### V.3. PROTOCOL

- Contaminated surface controls: Prepare 10 x 100 cm<sup>2</sup> of contaminated surfaces. These surfaces are wiped by sterile wipes used to microbial takings which are used in the laboratory. These controls allow to check the level of contamination.
- Sterile surface controls: Prepare 10 x 100 cm<sup>2</sup> of non contaminated surfaces. These surfaces are wiped by sterile wipes used to microbial takings which are used in the laboratory. These controls allow to check the sterility.
- Clean each contaminated surface with E cloth general purpose and water
- Rinse out the e-cloth with warm water
- Wipe, with the washed E-Cloth, a non contaminated surface (the same cloth)
- Measure bacteria left by the cloth on the non contaminated surface

## VI. RESULTS FOR EVALUATION OF BACTERIA TRANSFER

The results of contaminated and sterile surface controls are shown in the table below:

<b>Contaminated surface controls</b>	<b>Result (cfu/100cm<sup>2</sup>)</b>	<b>Sterile surface controls</b>	<b>Result (cfu/100cm<sup>2</sup>)</b>
Control 1	<10	Control 1	<10
Control 2	55 000	Control 2	<10
Control 3	<10	Control 3	<10
Control 4	~10	Control 4	<10
Control 5	~20	Control 5	<10
Control 6	1 100	Control 6	<10
Control 7	180 000	Control 7	<10
Control 8	1 300	Control 8	<10
Control 9	800	Control 9	<10
Control 10	35 000	Control 10	<10

These results show that the 10 sterile surfaces controls are sterile and the 10 contaminated surfaces are contaminated by *Escherichia coli*.

<b>Protocols</b>	<b>Additives</b>	<b>Results (cfu/100cm<sup>2</sup>)</b>	<b>% of bacteria transfer</b>
E Cloth general purpose	Water	<10	0.01
E Cloth general purpose	Water	<10	0.01
E Cloth general purpose	Water	~20	0.01
E Cloth general purpose	Water	<10	0.01
E Cloth general purpose	Water	<10	0.01
E Cloth general purpose	Water	<10	0.01
E Cloth general purpose	Water	~20	0.01
E Cloth general purpose	Water	~30	0.02
E Cloth general purpose	Water	~10	0.01
E Cloth general purpose	Water	<10	0.01

In order to calculate the percentage of bacteria transfer, 180 000 cfu/100cm<sup>2</sup> (control 7) is used.

#### Interpretation:

The results show that when the same cloth is used again (after simply washing under warm tap water), bacteria transfer is very low, about 0.01%.

## VII. CONCLUSION

### Evaluation of transfer bacteria

The evaluation of bacteria transfer show that when the same cloth is used again (after simply washing under warm tap water) bacteria transfer is very low, about 0.01%. E-Cloths remove bacteria, and the transfer of bacteria from the E-Cloth to other surface is only 0.01%.