CLIENT: Eruslu Saglik Urunleri San. Ve Tic. A.S.

ADDRESS: Baspinar (Organize) Osb Mh. O.S.B. 4. Bolge 83424 No'lu Cd. Eruslu Saglik Urunleri Sit. No:3 Sehitkamil / Gaziantep / TURKEY

Samples: Antibacterial Wet Wipes And Antibacterial Pocket Wipes

Sample received on: 22.05.2020

Report Number: 2020168090

Date Of Issue: 05.06.2020

Brands of the Product: Sleepy, Yess Baby, Purewipes , Penguin, Baby Turco, Soft Touch, Bioderminy, Mooncare, G&Y, Sweet Baby, Freshlife, Actual, Virowall, V-TUF

Tested by: Michael jones

Checked by: Jennifer Richardson

Rabi-Laboratories

Signed by: Walter Lee

Test Standard: EN 1275:2006

Test Purpose

Quantitative suspension test for the evaluation of basic fungicidal or basic yeasticidal activity of chemical disinfectants and antiseptics.

Test Conditions

The standard refers to the parameters to be observed when testing products for basic fungicidal or basic yeasticidal activity. This includes the test microorganism, test temperature and contact time.

Test microorganism refers to the mandatory list of microbes that must be used in the test to determine the antimicrobial activity of the product. The mandatory microorganisms are assumed to represent all microbes in its group.
Test temperature refers to the temperature in which the test must be conducted. The general assumption is that disinfectants are less effective in low temperatures compared to higher temperatures.
Contact time refers to the minimum duration a product must remain in contact with the microbes for the product to be effective.
One of the major differences between phase 1 basic suspension tests and phase 2 step 2 suspension tests is that phase 1 basic suspension tests do not include interfering substances in the test procedure.

Refer to the table below for the minimum test conditions for EN 1275.

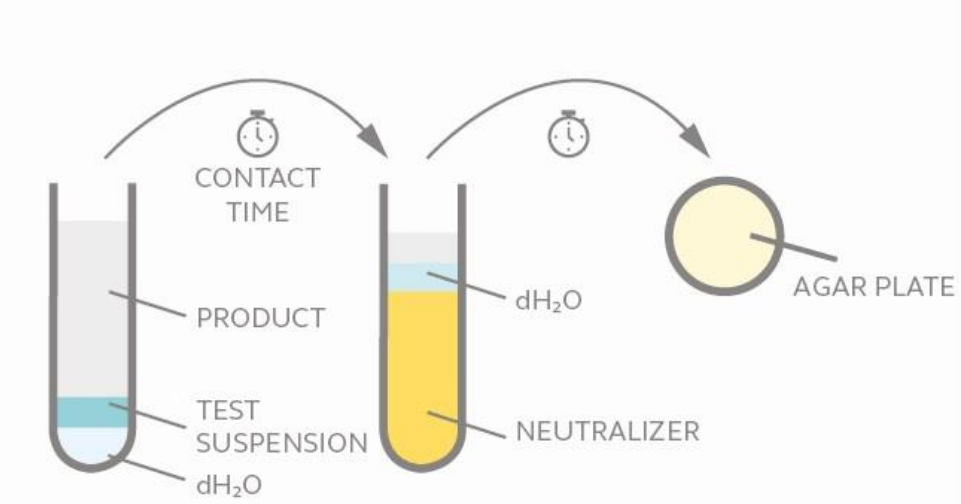
Table 1: EN 1275 test conditions and requirements

	Basic fungicidal or yeasticidal activity
Mandatory test microorganisms	Yeasticidal activity *C. albicans* Fungicidal activity *C. albicans* *A. brasiliensis*
Test temperature	20°C ± 1°C
Contact time	30 sec ± 10 sec
Reduction (lg)	≥ 4

Test Method

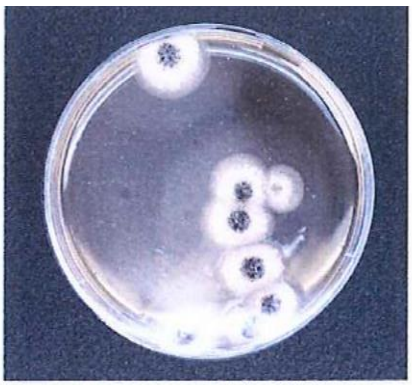
In a phase 1 suspension test, 8 parts of the test product is added to 1-part test microorganism and 1-part water. The mixture is allowed to interact for the duration of the contact time. One part of the mixture is added to 8 parts of neutralizer and 1-part water for 5 seconds to halt fungicidal or yeasticidal activity. The final mixture is then acquired and incubated for 3 days to allow surviving yeasts or fungi (if any) to proliferate. The yeasticidal or fungicidal colony is counted and compared against the original culture size.

Diagram 1: EN 1275 test method



Log Reduction

In order for this product to pass en 1275, yeast obtained a 4-day reduction against the relevant test microorganisms listed in Table 1 for lethal and fungicidal activities. In other words, the product can kill 99.99% yeast or fungi while meeting all other requirements of the European standard.



The test product passed the EN 1275.

Tested by: Michael jones

Checked by: Jennifer Richardson

Signed by: Walter Lee

Test Standard: EN 1650+A1:2013

Test Purpose

Quantitative suspension test for the evaluation of fungicidal or yeasticidal activity of chemical disinfectants and antiseptics used in food, industrial, domestic and institutional areas.
EN 1650 is a phase 2 step 1 suspension test for disinfectants intended for use in food, industrial, domestic and institutional areas. The test evaluates the efficacy of the product against fungi and yeasts.
Yeast and fungi occur naturally in the environment. Although many assume both microorganisms are the same, they are not. Fungus is a multi-celled microorganism that reproduces through spores. Yeast is a form of fungus but it is single-celled and it reproduces through mitosis. Mould is another form of fungi. Efficacy against mould is a mandatory requirement according to the European Norms.

Test Conditions

The standard refers to the parameters to be observed when testing products for general disinfection in the food, industrial, domestic and institutional areas. This includes the test microorganism, test temperature, contact time and interfering substance.

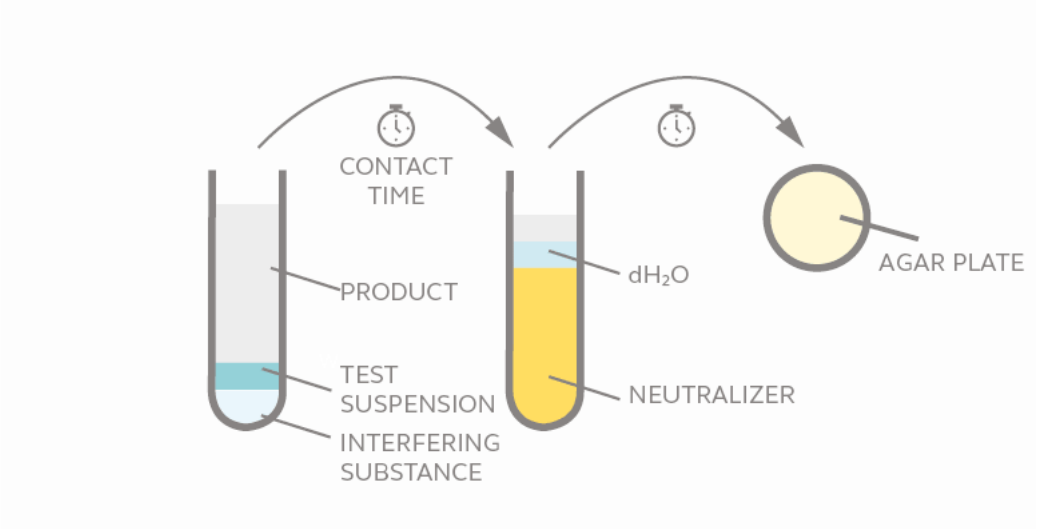
Test microorganism refers to the mandatory list of microbes that must be used in the test to determine the antimicrobial activity of the product. The mandatory microorganisms are assumed to represent all microbes in its group.
Test temperature refers to the temperature in which the test must be conducted. The general assumption is that disinfectants are less effective in low temperatures compared to higher temperatures.
Contact time refers to the minimum duration a product must remain in contact with the microbes for the product to be effective.
Interfering substance refers to substance used in the test to simulate the possible contaminants that co-occur with microbes in the actual environment.

Refer to the table below for the minimum and additional test conditions.

Table 1: EN 1650 test conditions and requirements

	General disinfection
Mandatory test microorganisms	Yeasticidal activity *C. albicans* Fungicidal activity *C. albicans* *A. brasiliensis*
Test temperature	20°C ± 1°C
Contact time	30 sec
Interfering substance – clean condition	0.3 g/l bovine albumin solution and /or
Interfering substance – dirty condition	3g/l bovine albumin solution
Reduction (lg)	≥ 4
Additional	Any relevant substance

Diagram 1: EN 1650 test method



Log Reduction

Log reduction refers to the extent to which a product is capable of reducing the number of microbes. For example, 4-log reduction means the number of microorganisms on a surface has been reduced by 10 000 times. A product that is 99.9% effective against a certain microbe is said to have achieved 3-log reduction against that microbe.

This product passed EN 1650 by achieving a 4-day reduction against the relevant test microorganisms listed in Table 1. In other words, the product can kill 99.99% yeast or mold while meeting all other requirements of the European standard.

The test product passed the EN 1650.

Tested by: Michael jones

Checked by: Jennifer Richardson

Signed by: Walter Lee

Rabi-Laboratories

Test Standard: EN 13624:2014

Test Purpose

Quantitative suspension test for the evaluation of fungicidal and yeasticidal activities of disinfectants intended for use in the medical area.

EN 13624 is a phase 2 step 1 suspension test performed on disinfectants intended for use in the medical area to test the efficacy of the product against fungi and yeasts.

Yeast and fungi occur naturally in the environment. Although many assume both microorganisms are the same, they are not. Yeast is a single-celled microorganism and it reproduces through mitosis. Fungus however, is a multi-celled microorganism that reproduces through spores. Efficacy against yeast is a mandatory requirement according to the European Norms.

Test Conditions

The standard refers to the parameters to be observed when testing products intended for use in the medical area. The parameter includes the test microorganism, test temperature, contact time and interfering substance.

Test microorganism refers to the mandatory list of microbes that must be used in the test to determine the antimicrobial activity of the product. The mandatory microorganisms are assumed to represent all microbes in its group.
Test temperature refers to the temperature in which the test must be conducted. The general assumption is that disinfectants are less effective in low temperatures compared to higher temperatures.

Contact time refers to the minimum duration a product must remain in contact with the microbes for the product to be effective.
Interfering substance refers to substance used in the test to simulate the possible contaminants that co-occur with microbes in the actual environment. In a hospital for example, hepatitis B virus contamination might also contain blood and other bodily fluids which may make disinfection challenging especially if it's dried matter.

Refer to the table below for the minimum EN 13624 test conditions.

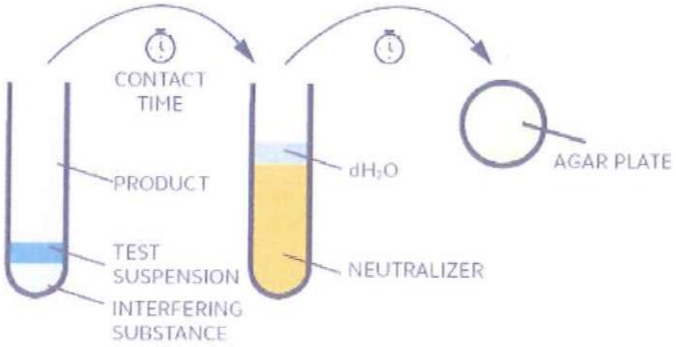
Table 1: EN 13624 test conditions and requirements

	Hygienic handrub and handwash	Surgical handrub and handwash	Instrument disinfection	Surface disinfection
Mandatory test microorganisms	*C. albicans* (vegetative cells)	*C. albicans* (vegetative cells)	Fungicidal activity *A. brasiliensis* *C. albicans* (vegetative cells) Yeasticidal activity *C. albicans* (vegetative cells)	Fungicidal activity *A. brasiliensis* *C. albicans* (vegetative cells) Yeasticidal activity *C. albicans* (vegetative cells)
Test temperature	According to the manufacturer's recommendation, but between			
	20°C and 20°C	20°C and 20°C	20°C and 70°C	4°C and 30°C
Contact time	According to the manufacturer's recommendation			
	but between		but no longer than	
	30 sec	1 min	30 min	5 min
Interfering substance – clean condition	0.3 g/l bovine albumin solution (hygienic handrub)	0.3 g/l bovine albumin solution (surgical handrub)	0.3 g/l bovine albumin solution and /or	0.3 g/l bovine albumin solution and /or
Interfering substance – dirty condition	0.3 g/l bovine albumin solution plus 3.0ml/l erythrocytes (hygienic handwash)	0.3 g/l bovine albumin solution plus 3.0ml/l erythrocytes (surgical handwash)	0.3 g/l bovine albumin solution plus 3.0ml/l erythrocytes	0.3 g/l bovine albumin solution plus 3.0ml/l erythrocytes
Reduction (lg)	≥ 4.0 for handrub products ≥ 2.0 for handwash products	≥ 4.0	≥ 4.0	≥ 4.0
Additional	Clean or dirty; any relevant substance	Clean or dirty; any relevant substance	Any relevant substance	Any relevant substance

Test Method

In EN 13624 phase 2 step 1 suspension test, 8 parts of the test product is added to 1-part test microorganism and 1-part interfering substance. The mixture is allowed to interact for the duration of the contact time. One part of the mixture is added to 8 parts of neutralizer and 1-part water for 5 minutes to halt fungicidal or yeasticidal activity. The final mixture is then acquired and incubated for 3 days to allow surviving yeast or fungi (if any) to proliferate. The colony is counted and compared against the original culture size.

Diagram 1: EN 13624 test method

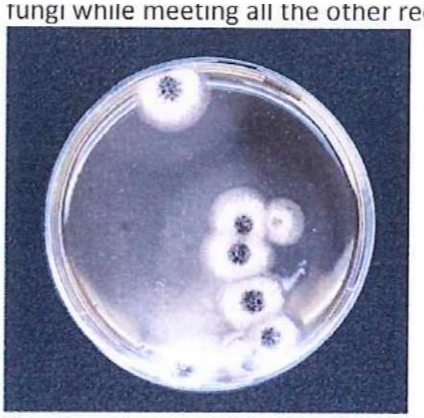


Log Reduction

Log reduction refers to the extent to which a product is capable of reducing the number of microbes. For example, 4-log reduction means the number of microorganisms on a surface has been reduced by 10 000 times. A product that is 99.9% effective against a certain microbe is said to have achieved

This product has passed EN 13624, by achieving 4-log reduction against the relevant test microorganisms listed in Table 1. In other words, the product kills 99.99% of yeasts or fungi while meeting all other requirements of the European standard.

fungi while meeting all the other requirements of the European standard.



The test product passed the EN 13624: 2014

Tested by: Michael jones

Checked by: Jennifer Richardson

Signed by: Walter Lee

Rabi-Laboratories

Test Standard: EN 1276:2009

Test Purpose

Quantitative suspension test for the evaluation of bactericidal activity of chemical disinfectants and antiseptics used in food, industrial, domestic and institutional areas.

EN 1276 is a phase 2 step 1 suspension test for disinfectants intended for use in food, industrial, domestic and institutional areas. The test evaluates the efficacy of the product against bacteria.

Test Conditions

The standard refers to the parameters to be observed when testing products for general disinfection in the food, industrial, domestic and institutional areas. This includes the test microorganism, test temperature, contact time and interfering substance.

Test microorganism refers to the mandatory list of microbes that must be used in the test to determine the antimicrobial activity of the product. The mandatory microorganisms are assumed to represent all microbes in its group.

Test temperature refers to the temperature in which the test must be conducted. The general assumption is that disinfectants are less effective in low temperatures compared to higher temperatures.

Contact time refers to the minimum duration a product must remain in contact with the microbes for the product to be effective.

Interfering substance refers to substance used in the test to simulate the possible contaminants that co-occur with microbes in the actual environment.

Refer to the table below for the minimum and additional test conditions.

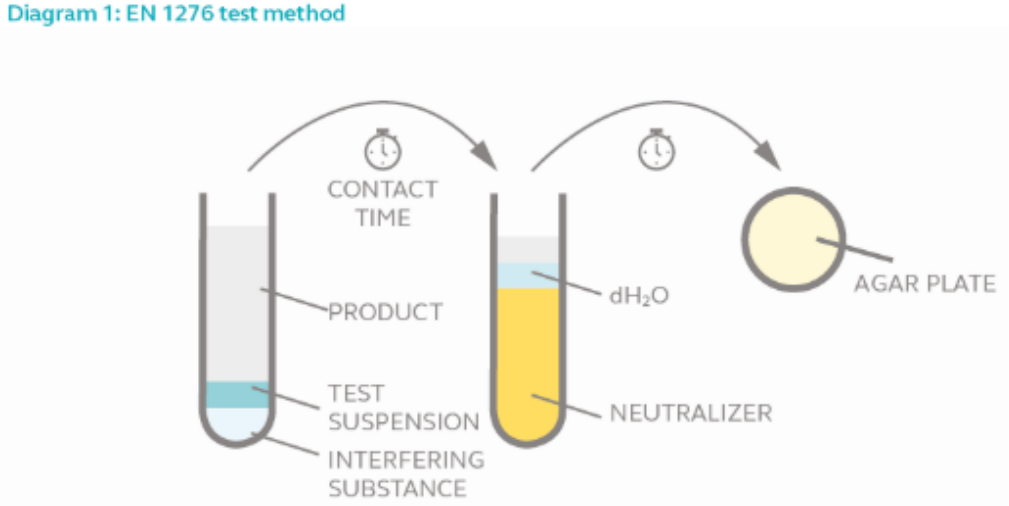
Table 1: EN 1276 test conditions and requirements

	General disinfection
Mandatory test microorganisms	*P. aeruginosa* *E. coli* *S. aureus* *E. hirae*
Test temperature	20°C ± 1°C
Contact time	5 min ± 10 sec 1 min (for hand disinfection)
Interfering substance – clean condition	0.3 g/l bovine albumin solution and /or
Interfering substance – dirty condition	3 g/l bovine albumin solution
Reduction (lg)	≥ 5
Additional	Any relevant substance

Test Method

In a phase 2 step 1 suspension test, 8 parts of the test product is added to 1-part test microorganism and 1-part interfering substance. The mixture is allowed to interact for the duration of the contact time. One part of the mixture is added to 8 parts of neutralizer and 1-part water for 5 minutes to halt bactericidal activity. The final mixture is then acquired and incubated for 2 days to allow surviving bacteria (if any) to proliferate. The bacterial colony is counted and compared against the original culture size.

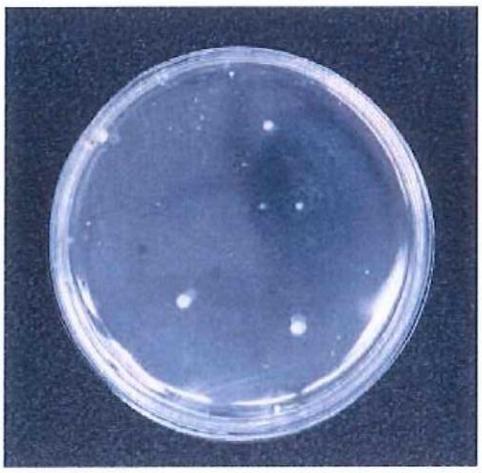
Diagram 1: EN 1276 test method



Log Reduction

Log reduction refers to the extent to which a product is capable of reducing the number of microbes. For example, 4-log reduction means the number of microorganisms on a surface has been reduced by 10 000 times. A product that is 99.99% effective against a certain microbe is said to have achieved 3-log reduction against that microbe.

This product has passed en 1276, by achieving a 5-day reduction against the relevant test microorganisms listed in Table 1. In other words, the product can kill 99.99% bacteria while meeting all other requirements of the European standard.



The test product passed the EN 1276:2009

Tested by: Michael jones

Checked by: Jennifer Richardson

Signed by: Walter Lee

Rabi-Laboratories

Test Standard: 13727+A2: 2015

Test Purpose

Quantitative suspension test for the evaluation of bactericidal activity of disinfectants intended for use in the medical area.

EN 13727+A2:2015 is a mandatory phase 2 step 1 suspension test for disinfectants intended for use in the medical area. The test evaluates the efficacy of the product against bacteria.

Test Conditions

The standard refers to the parameters to be observed when testing products intended for surface disinfection, instrument disinfection, surgical and hygienic handrub and handwash. This includes the test microorganism, test temperature, contact time and interfering substance.

Test microorganism refers to the mandatory list of microbes that must be used in the test to determine the antimicrobial activity of the product. The mandatory microorganisms are assumed to represent all microbes in its group.

Test temperature refers to the temperature in which the test must be conducted. The general assumption is that disinfectants are less effective in low temperatures compared to higher temperatures.

Contact time refers to the minimum duration a product must remain in contact with the microbes for the product to be effective.

Interfering substance refers to substance used in the test to simulate the possible contaminants that co-occur with microbes in the actual environment. In a hospital for example, hepatitis B virus contamination might also contain blood and other bodily fluids which may make disinfection challenging especially if it's dried matter.

Refer to the table below for the minimum and additional test conditions.

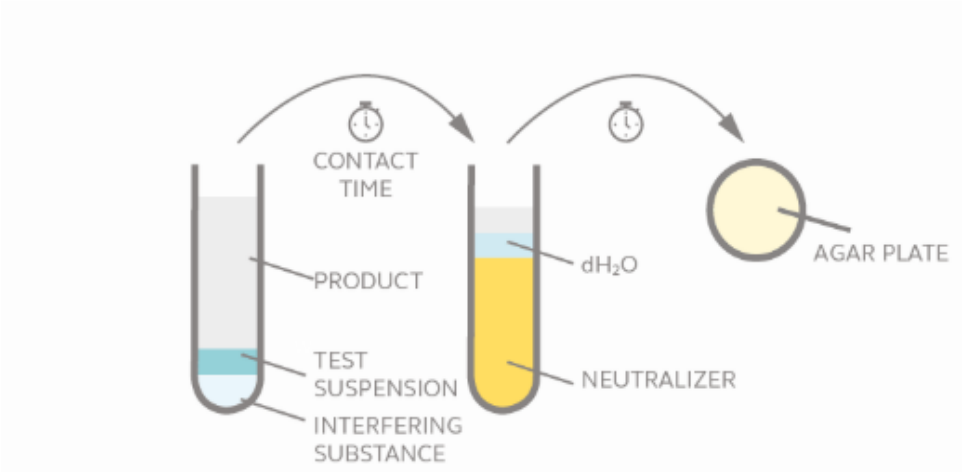
Table 1: EN 13727 test conditions and requirements

	Hygienic handrub and handwash	Surgical handrub and handwash	Instrument disinfection	Surface disinfection
Mandatory test microorganisms	*P. aeruginosa* *S. aureus* *E. hirae* *E. coli K12*	*P. aeruginosa* *S. aureus* *E. hirae* *E. coli K12*	*P. aeruginosa* *S. aureus* *E. hirae* When temperature is 40°C or higher: only *E. faecium*	*P. aeruginosa* *S. aureus* *E. hirae*
Test temperature	According to the manufacturer's recommendation, but between			
	20°C and 20°C	20°C and 20°C	20°C and 70°C	4°C and 30°C
Contact time	According to the manufacturer's recommendation			
	but between		but no longer than	
	30 sec and 60 sec	1 min and 5 min	60 min	5 min or 60 min
Interfering substance – clean condition	0.3 g/l bovine albumin solution (hygienic handrub)	0.3 g/l bovine albumin solution (surgical handrub)	0.3 g/l bovine albumin solution and /or	0.3 g/l bovine albumin solution and /or
Interfering substance – dirty condition	0.3 g/l bovine albumin solution plus 3.0ml/l erythrocytes (hygienic handwash)	0.3 g/l bovine albumin solution plus 3.0ml/l erythrocytes (surgical handwash)	0.3 g/l bovine albumin solution plus 3.0ml/l erythrocytes	0.3 g/l bovine albumin solution plus 3.0ml/l erythrocytes
Reduction (lg)	≥ 5.0 for handrub products ≥ 3.0 for handwash products	≥ 5.0	≥ 5.0	≥ 5.0
Additional	Clean or dirty; any relevant substance	Clean or dirty; any relevant substance	Any relevant substance	Any relevant substance

Test Method

In a phase 2 step 1 suspension test, 8 parts of the test product is added to 1-part test microorganism and 1-part interfering substance. The mixture is allowed to interact for the duration of the contact time. One part of the mixture is added to 8 parts of neutralizer and 1-part water for 5 minutes to halt bactericidal activity. The final mixture is then acquired and incubated for 2 days to allow surviving bacteria (if any) to proliferate. The bacterial colony is counted and compared against the original culture size.

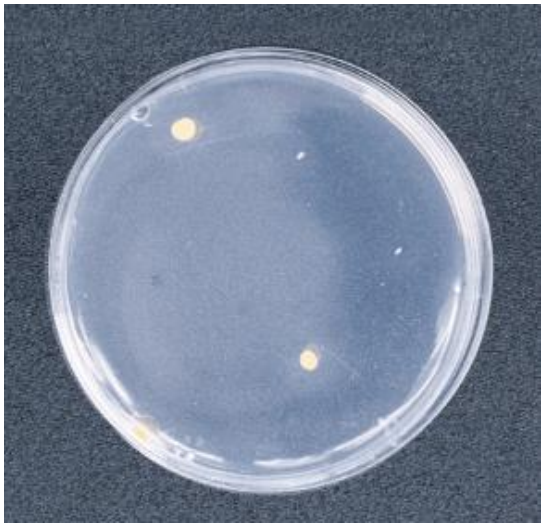
Diagram 1: EN 13727 test method



Log Reduction

Log reduction refers to the extent to which a product is capable of reducing the number of microbes. For example, 4-log reduction means the number of microorganisms on a surface has been reduced by 10 000 times. A product that is 99.9% effective against a certain microbe is said to have achieved 3-log reduction against that microbe.

This product has passed EN 13727, by achieving a 5-day reduction against the relevant test microorganisms listed in Table 1. In other words, the product kills 99.99% bacteria while meeting all other requirements of the European standard.



The test product passed the EN 13727:2012+A2: 2015

Tested by: Michael jones

Checked by: Jennifer Richardson

Signed by: Walter Lee

Rabi-Laboratories

Test Standard: EN 14476+A1:2015, EN 14476+A2:2019

Test Purpose

Quantitative suspension test for the evaluation of virucidal activity of disinfectants intended for use in the medical area.

EN 14476 is a phase 2 step 1 suspension test to evaluate the virucidal activity of chemical disinfectants intended for use in the medical area.

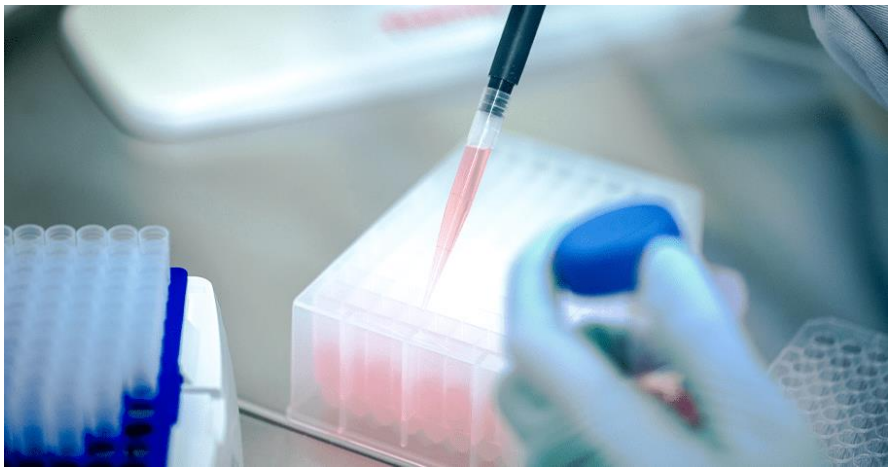
Refer to the table below for the minimum requirements for EN 14476+A1+A2 test:

Table 1: EN 14476 mandatory test viruses

	Hygienic handrub and handwash	Instrument disinfection	Surface disinfection	Textile disinfection
Mandatory test microorganisms	Fully virucidal Adenovirus Norovirus Poliovirus Limited spectrum virucidal activity Adenovirus Norovirus Virucidal activity against enveloped viruses Vaccinia virus	Adenovirus Norovirus When temperature is 40°C or higher, only parvovirus	Adenovirus Norovirus Poliovirus	Parvovirus
Test temperature	according to the manufacturer's recommendation but at / between			
	20°C	20°C and 70°C	4°C and 30°C	30°C and 70°C
Contact time	according to the manufacturer's recommendation			
	but between	but no longer than	but no longer than	but no longer than
	30 sec and 2 min	60 min	5 min or 60 min	20 min
Interfering substance – clean condition	0,3 g/l bovine albumin solution (hygienic handrub)	0,3 g/l bovine albumin solution and/or	0,3 g/l bovine albumin solution and/or	
Interfering substance – dirty condition	3,0 g/l bovine albumin solution plus 3,0 ml/l erythrocytes (hygienic handwash)	3,0 g/l bovine albumin solution plus 3,0 ml/l erythrocytes	3,0 g/l bovine albumin solution plus 3,0 ml/l erythrocytes	3,0 g/l bovine albumin solution plus 3,0 ml/l erythrocytes
Reduction (lg)	≥ 4	≥ 4	≥ 4	≥ 4

Test Conditions

All the viruses listed are non-enveloped viruses (except vaccinia virus) and as such are more resistant to chemical disinfectants compared to enveloped viruses. Poliovirus is the most resistant among non-enveloped viruses and manufacturers often struggle to attain efficacy against this microorganism. If EN 14476 is performed against all three non-enveloped viruses above for a hand product and it passes the test for adenovirus or norovirus but fails against poliovirus, the product is deemed as limited spectrum virucidal or limited virucidal. For a hand product to be fully virucidal or acknowledged as capable of inactivating all enveloped and non-enveloped viruses, it must be effective against adenovirus, norovirus and poliovirus. Instrument and surface disinfectants intended for the medical area however, must pass the test against all three non-enveloped viruses.



In preparing for the suspension test, the test virus is added to an interfering substance in a suspension. The choice of interfering substance used in the test depends on the product claim. The test product or disinfectant is then added to the virus suspension for the duration of the exposure time at the temperature specified by the manufacturer.

At the end of the exposure time, samples are retrieved and the activity of the test product is neutralised by dilution in ice-cold test medium. Serial dilutions are performed, and the dilutions are examined for viral infectivity.

Unlike bacteria or fungus, most viruses are too small (ranging from 25nm to 400nm) for observation under a light microscope. The presence of viruses in a suspension before and after product exposure is therefore determined by inoculating live host cells with suspension samples. These cells are then observed after 7 days (depending on the cell type) for structural changes. If the test product had not been successful in inactivating test viruses before neutralization, they invade and damage the live cells to display cytopathic effect (CPE). These are the effects virologists look for when observing the cells under a light microscope.

Control Tests

In addition to the efficacy test, 5 control tests are run concurrently to eliminate other possible explanations for the test results. The 5 control tests are:

1. Virus control

Determines the infectivity of the test virus suspension. To pass the test, the concentration of virus in the control test must be sufficiently high to enable a 4-log reduction or reduction of viruses by 10 000-fold.

2. Cytotoxicity control

Reveals the possible alteration in cell structure caused by the test product or disinfectant. To pass the test, live cells must not display toxic reaction or damage to a level where achieving 4-log reduction is not possible.

3. Suppression control

Verifies the efficiency of the neutralising method in suppressing the virucidal activity of the test product after the required exposure time.

4. Interference control

Verifies the susceptibility of infection in cells is not influenced negatively by the test product (passed cytotoxicity test).

Ensures the test virus can be inactivated and non-resistant to antimicrobial agents, enabling it to achieve 4-log reduction.

Influenza A (H1N1)

	Refrance of virus	% 0.1 Product Effect			
		30 Seconds		60 Seconds	
Virus Titre*	5.5	**Clean Environment**	**Dirty Environment**	**Clean Environment**	**Dirty Environment**
Virus Titre With Product**		1.5	1.5	1.3	1.5
The Rate Of Decrease In The Titre Of The Virus***		4.0	4.0	4.2	4.0

* Logarithmic TCID50 value of virus in M1.
**Logarithmic TCID50 value of the virus treated with the product in different time periods and environments.
***Logarithmic TCID50 ratio between virus titer and product virus titer.

It tested the effectiveness of a liquid wet cloth solution of Eruslu Saglik Urunleri San. Ve Tic. A.S. against the influenza A (H1N1) virus in accordance with EN 14476 standards. The lowest non-toxic rate of the disinfectant solution in question of the product tested in this experiment was used in this study because the 100% ratio showed toxic effects to cells in the test environment. As a result of the Test, the product at 0.1% pda temperature (20 C), in clean and dirty conditions, 30 and 60 seconds of application time, in all experimental conditions virus titer (see result table) was found to cause at least 4 log reductions. According to Antimicrobial Division US EPA standards, disinfectants are required to reduce virus titers by 4 logs or more for their virusidal activity.

As a result; the results of this experiment show that the product tested was 99.99% effective against the influenza A (H1N1) virus at the 0.1% suspension of 30 and 60 seconds of application time.

This product can maintain its effectiveness if used with one of the methods of washing, wiping, impregnation (wetting/dipping) and spraying, provided that it is used at least in the above-mentioned resolutions and times.

Coronavirus (COVID-19)

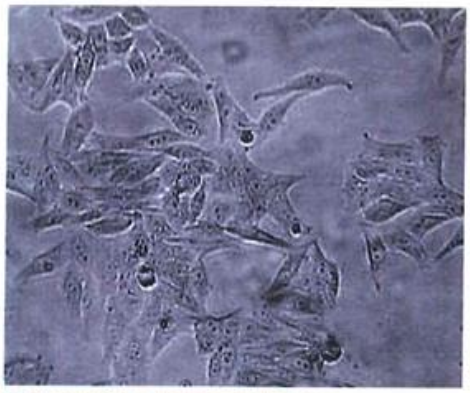
It tested the effectiveness of a liquid wet cloth solution of Eruslu Saglik Urunleri San. Ve Tic. A.S. against the coronavirus (COVID-19) in accordance with EN 14476 standards. The lowest non-toxic rate of the disinfectant solution in question of the product tested in this experiment was used in this study because the 100% ratio showed toxic effects to cells in the test environment. As a result of the Test, the product at 0.1% pda temperature (20 C), in clean and dirty conditions, 5 and 30 seconds of application time, in all experimental conditions virus titer (see result table) was found to cause at least 4 log reductions. According to Antimicrobial Division US EPA standards, disinfectants are required to reduce virus titers by 4 logs or more for their virusidal activity.

As a result; the results of this experiment show that the product tested was 99.99% effective against the coronavirus (COVID-19) at the 0.1% suspension of 30 seconds of application time.

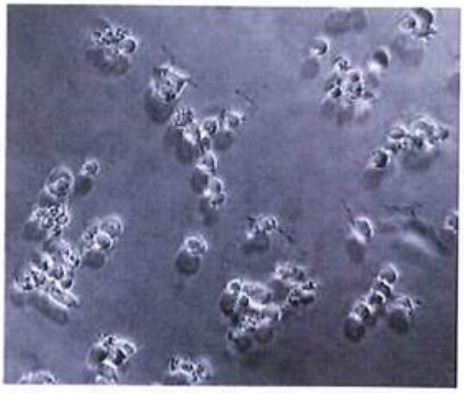
This product may retain its effectiveness when used in one of the methods of washing, wiping, impregnation (wetting/dipping) and spraying, provided it is used at specified resolutions and times.

Determination of Results

CPEs if present, can be viewed through a light microscope and the reduction in virus infectivity is calculated through the difference in virus concentration before and after treatment with the test product. A 4-log reduction or reduction of viruses by 10 000-fold demonstrates the ability of the test product in inactivating viruses to a level acceptable to the European standards. Log reductions are calculated by determining 50% Tissue Culture Infective Dose (TCID50) or the viral dose required to display CPE in 50% of the cell culture.



Cell lines before exposure to Poliovirus



Cell lines displaying CPE after exposure to Poliovirus

Tested by: Michael jones

Checked by: Jennifer Richardson

Signed by: Walter Lee

Rabi-Laboratories

Test Standard: EN 16615:2015

Test Purpose

EN 16615:2015 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)

Medical Area – Standard Test Methods To Be Used To Substantiate Claims For Products

Type of Activity	Phase, step	Product Claim / Field of Application					
		Hygienic Handrub	Hygienic Handwash	Surgical Handrub or -wash	Surface Disinfection		Instrument Disinfection
					mechanical action		
					without	with	
Bactericidal	2,1	EN 13727 (handrub products under clean, handwash products under dirty conditions)			EN 13727		EN 13727
	2,2	EN 1500	EN 1499	EN 12791	EN 13697	EN 16615	EN 14561
Yeasticidal	2,1	EN 13624 (handrub products under clean, handwash products under dirty conditions)			EN 13624		EN 13624
	2,2	*No tests available*			EN 13697	EN 16615	EN 14562

The method

A strip of PVC with polyurethane surface coating measuring 20 x 50 cm is prepared by outlining four 5 x 5 cm squares to reflect the test area / test field.

The first square is inoculated with 50 ml of either bacteria or yeast.

The inoculum is spread evenly on the first square and left to dry. The rest of the squares are left untouched.

A test wipe is placed just before the inoculated square and a 2.5 kg weight is positioned on the wipe. The weight is to imitate the pressure applied by hand when wiping. The weight also ensures that only the amount released onto the surface is available to disinfect.

The weight is pushed from the side across the first square all the way to the fourth square in a swift 2-second motion to replicate physical wiping action.

The weight is then pushed back across all the squares to the first square in another swift 2-second motion.

The suggested contact time is observed before each square is sampled for observation.

An alternative method outlined in the standard suggests using a dry wipe recommended in the norm (or supplied by the manufacturer) soaked in 16 ml of disinfectant solution for 30 minutes. The wipe then goes through the same steps as above.

To pass, the test must eliminate 99.999% of bacteria on the surface (5-log reduction) or eliminate 99.99% of yeasts (4-log reduction).

This product has passed EN 16615, by achieving a 5-day reduction against the relevant test microorganisms. In other words, the product kills 99.99% bacteria while meeting all other requirements of the European standard.

Tested by: Michael jones

Checked by: Jennifer Richardson

Signed by: Walter Lee

Test Standard: EN 1500: 2013

Test Purpose

Chemical disinfectants and antiseptics - Hygienic handrub - Test method and requirements (phase 2/step 2)

Test Method

This standard specifies method of test simulating practical conditions for establishing whether aproduct for hygienic handru bieduces the release of transient flora according to the requirements when rubbed an to the articfically contaminated hads of volunteers.

Its specifies a test method simulating practical conditions for establishing whether a product for hygienic handrub reduces the release of transient microbial flora on hands when rubbed onto the artificially contaminated hands of volunteers. NOTE 1 Attention is drawn to the fact that tests on human volunteers are the subject of legal provisions in certain European countries/regions. This European Standard applies to products for hygienic handrub for use in areas and situations where disinfection is medically indicated. Such indications occur in patient care, for example: - in hospitals, in community medical facilities and in dental institutions; - in clinics of schools, of kindergardens and of nursing homes. And may occur in the workplace and in the home. It may also include services such as laundries and kitchens supplying products directly for the patient. EN 14885 specifies in detail the relationship of the various tests to one another and to "use recommendations". NOTE 2 This method corresponds to a phase 2, step 2 test.

Experimental Conditions as outlined in the standard

Neutraliser: BU Broth

Bacterial Strains: E. coli K12 NCTC 10538

Product Dilution: Neat (as supplied)

Test Product Application: 4ml applied to dry hands. Hands rubbed for 60s.

Incubation Temperature: 36˚C ± 1˚C

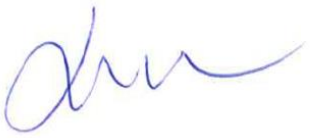
Conclusion

The mean pairwise differences that do not exceed the median (here -0.18) are computed. The critical values for Wilcoxon's matched pairs signed ranks test the entry for n=18 and a one-sided 0.025 level of significance, the critical value of 40 is found. Hence, c=40+1=41. The 41th entry is 0.13. Hence, the Hodges-Lehmann upper one sided 97.5% confidence limit for the difference in lg reductions between RP and PP is 0.13 which is less than the agreed inferiority margin of 0.6 lg units. Therefore the hypothesis of inferiority of PP (test product) is rejected and it can be concluded that the test preparation PP is not inferior to RP (reference product)

Results

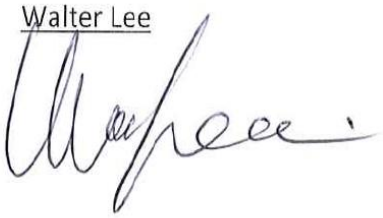
Subject	Hand	Mean Pre Value	Mean L+R	Mean Log Pre Value	Mean Post Value	Mean L+R	Mean Log Post Value	Log Reduction Factor
1	L	4.10E+06	3.00E+06	6.48	3.40E+03	2.85E+03	3.45	3.02
	R	1.90E+06			2.30E+03			
2	L	9.60E+05	8.40E+05	5.92	7.80E+02	8.25E+02	2.92	3.01
	R	7.20E+05			8.70E+02			
3	L	3.20E+06	3.25E+06	6.51	1.83E+03	2.42E+03	3.38	3.13
	R	3.30E+06			3.00E+03			
4	L	7.50E+05	7.40E+05	5.87	3.10E+01	2.06E+02	2.31	3.56
	R	7.30E+05			3.80E+02			
5	L	6.20E+05	5.60E+05	5.75	5.60E+02	5.20E+02	2.72	3.03
	R	5.00E+05			4.80E+02			
6	L	6.20E+05	1.32E+06	6.12	8.30E+02	6.25E+02	2.80	3.32
	R	2.01E+06			4.20E+02			
7	L	6.00E+05	5.45E+05	5.74	1.61E+02	2.36E+02	2.37	3.36
	R	4.90E+05			3.10E+02			
8	L	8.10E+06	7.95E+06	6.90	1.44E+03	9.65E+02	2.98	3.92
	R	7.80E+06			4.90E+02			
9	L	4.60E+06	4.25E+06	6.63	3.10E+03	3.80E+03	3.58	3.05
	R	3.90E+06			4.50E+03			
10	L	1.53E+06	1.56E+06	6.19	9.00E+02	9.00E+02	2.95	3.24
	R	1.58E+06			9.00E+02			
11	L	1.21E+06	1.03E+06	6.01	4.00E+02	6.20E+02	2.79	3.22
	R	8.40E+05			8.40E+02			
12	L	5.70E+05	4.25E+05	5.63	3.70E+02	2.80E+02	2.45	3.18
	R	2.80E+05			1.90E+02			
13	L	1.77E+05	1.78E+05	5.25	2.20E+02	2.43E+02	2.38	2.87
	R	1.79E+05			2.65E+02			
14	L	2.48E+06	2.02E+06	6.30	2.78E+03	2.61E+03	3.42	2.89
	R	1.55E+06			2.43E+03			
15	L	4.60E+06	5.45E+06	6.74	7.00E+02	1.42E+03	3.15	3.59
	R	6.30E+06			2.13E+03			
16	L	1.40E+05	2.90E+05	5.46	4.10E+02	1.14E+03	3.05	2.41
	R	4.40E+05			1.86E+03			
17	L	2.90E+06	3.70E+06	6.57	4.30E+02	4.75E+02	2.68	3.89
	R	4.50E+06			5.20E+02			
18	L	3.60E+06	5.85E+06	6.77	5.30E+02	5.10E+02	2.71	4.06
	R	8.10E+06			4.90E+02			

Tested by: Michael jones



Checked by: Jennifer Richardson

Signed by: Walter Lee



Rabi-Laboratories