Nordic Tersus
Laboratory

Test report No. hd0618

HYGIENIC HANDRUB TEST (EN 1500)

Name of the product:	CHEMISEPT MED
Batch number:	196101017
Order number:	17029
Manufacturer:	Chemi-Pharm Ltd.
Client, representative:	Chemi-Pharm Ltd., Põllu 132, Tallinn, 10917, ESTONIA Maris Millner, +372-51-77-090
Date of delivery:	10.01.2018
Test material conditions:	No specific features, sample in the manufacturers tare
Storage conditions:	In room temperature, dark
Active substance – conc.:	Ethyl alcohol 72.5% wt, isopropyl alcohol 7.5% wt
Appearance of the product:	Transparent liquid
Test concentration:	Ready to use
Contact time:	30 s
Interfering substance:	-
Rinsing liquid:	-
Neutralizer:	Polysorbate 80, 30 g/l; saponin, 30 g/l; lecithin, 3 g/l
Test organisms:	*Escherichia coli* K12 NCTC 10538
Testing method based on:	EVS-EN 1500:2013 Chemical disinfectants and antiseptics - Hygienic handrub - Test method and requirements (phase 2/step 2)
Testing date:	01.02.2018 - 02.02.2018
Results:	look appendix 1 - 6

ACCREDITED LABORATORY No L263 ISO/IEC 17025 NORDIC TERSUS LABORATORY OÜ TALLINN ESTONIA

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Diana Kaare, MSc

Head of laboratory, microbiologist

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Date: 2023.01.06 14:20:03 EET
Reason: MoldSign Signature
Location: Moldova

Date of test report: 06.02.2018

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Appendix 1

Reference hygienic handrub – experimental results

Reference solution: [propan-2-ol alcohol 60% (v/v)]
Handrub procedure: 3 ml 30 sec. Repeat the procedure.
Date: 01.02.2018 - 02.02.2018
Test organism: *Escherichia coli* K12 NCTC 10538; Suspension: 2,19 x 10^8 cfu/ml

Volunteer			Number of CFU per plate from dilution 10^x					
			Prevalues			Postvalues		
No	Hand, left or right	Sequence	-3	-4	-5	0	-1	-2
1	l	PP>RP	>330	104	12	31	5	0
	r		>330	92	11	36	5	0
2	l	PP>RP	>330	48	7	23	2	0
	r		>330	63	10	28	5	0
3	l	PP>RP	>330	114	8	52	9	2
	r		>330	86	6	43	2	0
4	l	PP>RP	>330	51	5	27	6	2
	r		>330	37	5	21	4	0
5	l	PP>RP	306	35	5	15	1	0
	r		281	31	6	18	4	0
6	l	PP>RP	>330	41	7	27	5	0
	r		>330	72	13	33	6	2
7	l	PP>RP	>330	91	15	62	10	4
	r		>330	74	4	49	11	2
8	l	PP>RP	>330	57	9	41	8	0
	r		>330	83	14	55	4	0
9	l	RP>PP	>330	66	4	29	4	0
	r		>330	81	6	25	5	1
10	l	RP>PP	>330	94	16	39	4	0
	r		>330	77	13	29	1	0
11	l	RP>PP	>330	42	5	22	7	2
	r		>330	59	6	31	5	0
12	l	RP>PP	>330	63	13	28	2	0
	r		>330	85	14	40	9	4
13	l	RP>PP	>330	91	11	71	13	3
	r		>330	54	7	58	9	2
14	l	RP>PP	>330	121	20	77	15	3
	r		>330	106	15	52	10	2
15	l	RP>PP	>330	198	24	93	19	4
	r		>330	175	22	78	9	3
16	l	RP>PP	>330	159	23	64	5	1
	r		>330	177	19	70	10	3
17	l	RP>PP	>330	314	28	103	14	4
	r		>330	286	23	92	13	0
18	l	RP>PP	>330	141	19	80	11	0
	r		>330	124	17	63	4	1

55= count used for computations; 55= adjacent usage; >330 = not countabable

Appendix 2

Hygienic handrub procedure with the product under test – experimental results

Product: CHEMISEPT MED
Handrub procedure: 1x3 ml 30 sec
Date: 01.02.2018 - 02.02.2018
Test organism: *Escherichia coli* K12 NCTC 10538; Suspension: 2,19 x 10^8 cfu/ml

Volunteer			Number of CFU per plate from dilution 10^x					
			Prevalues			Postvalues		
No	Hand, left or right	Sequence	-3	-4	-5	0	-1	-2
1	l	PP>RP	>330	58	11	5	0	0
	r		>330	66	4	3	0	0
2	l	PP>RP	>330	81	10	11	2	0
	r		>330	119	11	14	3	0
3	l	PP>RP	>330	123	15	24	5	1
	r		>330	104	8	18	2	0
4	l	PP>RP	>330	>330	31	68	10	2
	r		>330	281	30	41	7	2
5	l	PP>RP	>330	94	10	23	4	0
	r		>330	73	9	28	5	0
6	l	PP>RP	>330	>330	47	51	7	0
	r		>330	>330	61	70	9	2
7	l	PP>RP	>330	101	13	9	2	0
	r		>330	85	10	14	2	0
8	l	PP>RP	>330	69	5	6	0	0
	r		>330	94	12	7	0	0
9	l	RP>PP	>330	121	12	17	4	0
	r		>330	139	18	12	2	0
10	l	RP>PP	>330	105	17	25	4	0
	r		>330	74	11	28	5	0
11	l	RP>PP	237	19	3	2	0	0
	r		281	25	5	6	0	0
12	l	RP>PP	>330	81	13	11	2	0
	r		>330	98	10	15	5	1
13	l	RP>PP	>330	57	9	17	3	0
	r		>330	73	9	23	0	0
14	l	RP>PP	>330	87	11	22	4	0
	r		>330	99	15	28	5	1
15	l	RP>PP	>330	224	26	48	7	1
	r		>330	191	23	34	5	0
16	l	RP>PP	>330	154	18	27	3	1
	r		>330	141	17	15	3	0
17	l	RP>PP	>330	>330	52	81	10	3
	r		>330	>330	38	70	11	4
18	l	RP>PP	>330	162	14	9	2	0
	r		>330	133	10	9	3	0

55 = count used for computations; 55 = adjacent usage; >330 = not countable

Appendix 3

List of computed lg values (means of left and right hands) and lg reductions

Volunteer	Sequence	Reference handrub (RP) (Propan-2-ol 60% V/V)			Handrub with product under test (pp)		
		lg prevalues	lg postvalues	lg R	lg prevalues	lg postvalues	lg R
1	PP>RP	5.99	2.52	3.47	5.79	1.59	4.20
2	PP>RP	5.74	2.40	3.34	5.99	2.09	3.90
3	PP>RP	6.00	2.67	3.32	6.05	2.32	3.74
4	PP>RP	5.64	2.38	3.26	6.47	2.72	3.75
5	PP>RP	5.47	2.22	3.25	5.92	2.40	3.52
6	PP>RP	5.74	2.47	3.26	6.73	2.78	3.95
7	PP>RP	5.91	2.74	3.17	5.97	2.05	3.92
8	PP>RP	5.84	2.68	3.16	5.91	1.81	4.09
9	PP>RP	5.86	2.43	3.43	6.11	2.15	3.96
10	RP>PP	5.93	2.53	3.40	5.95	2.42	3.52
11	RP>PP	5.70	2.42	3.28	5.41	1.54	3.87
12	RP>PP	5.86	2.52	3.34	5.95	2.11	3.84
13	RP>PP	5.85	2.81	3.04	5.81	2.30	3.51
14	RP>PP	6.05	2.80	3.25	5.97	2.39	3.57
15	RP>PP	6.27	2.93	3.34	6.32	2.61	3.71
16	RP>PP	6.22	2.83	3.40	6.17	2.31	3.86
17	RP>PP	6.48	2.99	3.49	6.65	2.88	3.77
18	RP>PP	6.12	2.85	3.27	6.17	1.95	4.21
X	Overall	5.93	2.62	3.30	6.07	2.25	3.83
s		0.24	0.22	0.11	0.32	0.38	0.22
NN		18	18	18	18	18	18
X	PP>RP	5.80	2.50	3.30	6.10	2.21	3.89
s		0.17	0.17	0.11	0.30	0.39	0.20
NN		9	9	9	9	9	9
X	RP>PP	6.05	2.74	3.31	6.04	2.28	3.76
s		0.25	0.20	0.13	0.34	0.38	0.22
NN		9	9	9	9	9	9

lg R = decimal log reduction
RP>PP Sequence: first RP, second PP
PP>RP Sequence: first PP, second RP
X = Mean
s = Standard deviation
NN = Number on values (=volunteers)

Difference of mean Rs (RP>PP): 3.31 – 3.76 = (-0.45);
Difference of mean Rs (PP>RP): 3.30 – 3.89 = (-0.59);
Absolute difference of differences: Abs [(-0.59) - (-0.45)] = 0.14

Check of acceptance criteria:

1) Complete set of results from 18 volunteers available (minimum 18)
2) Mean of lg prevalues for RP = 5.93 and for PP = 6.07 (hence both greater than 5.00)
3) Individual lg reductions less than 3.00: with RP = 0 and with PP = 0
4) For group with sequence RP>PP difference of lg R: 3.31 – 3.76 = (-0.45); for group with sequence PP>RP difference of lg R: 3.30 – 3.89 = (-0.59); absolute difference of mean differences: Abs [(-0.59) - (-0.45)] = 0.14 (hence less than 2.00)
5) All quotients of weighted mean counts between 5 and 15 (results which were used for weighted mean counts in appendix 1 and appendix 2)

Acceptance criteria are fulfilled.

Appendix 4

Computation of individual differences of lg Rs of RP - PP

Volunteer	lg reduction (R)		Difference RP-PP
	Reference procedure (RP)	Product procedure (PP)	
1	3.47	4.20	-0.74
2	3.34	3.90	-0.56
3	3.32	3.74	-0.41
4	3.26	3.75	-0.49
5	3.25	3.52	-0.26
6	3.26	3.95	-0.69
7	3.17	3.92	-0.74
8	3.16	4.09	-0.93
9	3.43	3.96	-0.52
10	3.40	3.52	-0.12
11	3.28	3.87	-0.59
12	3.34	3.84	-0.50
13	3.04	3.51	-0.48
14	3.25	3.57	-0.32
15	3.34	3.71	-0.37
16	3.40	3.86	-0.46
17	3.49	3.77	-0.28
18	3.27	4.21	-0.94

Appendix 5

Sorting of individual differences and computation for Hodges-Lehmann 97,5% upper confidence limits

No	Sorted differences	Mean pairwise differences (di+dii)/2							
		-0.12	-0.26	-0.28	-0.32	-0.37	-0.41	-0.46	-0.48
1	-0.12	-0.12/1							
2	-0.26	-0.19/2	-0.26/6						
3	-0.28	-0.20/3	-0.27/7	-0.28/9					
4	-0.32	-0.22/4	-0.29/11	-0.30/12	-0.32/17				
5	-0.37	-0.24/5	-0.31/16	-0.32/18	-0.34/22	-0.37/30			
6	-0.41	-0.27/8	-0.34/21	-0.35/23	-0.37/29	-0.39/37	-0.41		
7	-0.46	-0.29/10	-0.36/24	-0.37/28	-0.39/36	-0.42	-0.44	-0.46	
8	-0.48	-0.30/13	-0.37/26	-0.38/31	-0.40/39	-0.42	-0.44	-0.47	-0.48
9	**-0.49**	-0.30/14	-0.37/27	-0.38/33	-0.40/40	-0.43	-0.45	-0.47	-0.48
10	**-0.50**	-0.31/15	-0.38/31	-0.39/35	-0.41	-0.44	-0.46	-0.48	-0.49
11	-0.52	-0.32/19	-0.39/34	-0.40/38	-0.42	-0.45	-0.47	-0.49	-0.50
12	-0.56	-0.34/20	-0.41	-0.42	-0.44	-0.46	-0.48	-0.51	
13	-0.59	-0.36/25	-0.43	-0.44	-0.46	-0.48	-0.50		
14	-0.69	**-0.41/41**	-0.48	-0.49	-0.51	-0.53			
15	-0.74	-0.43	-0.50	-0.51	-0.53				
16	-0.74	-0.43	-0.50	-0.51					
17	-0.93	-0.53	-0.60						
18	-0.94	-0.53							

The differences of the individual lg R's of RP-PP from table at the Appendix 4 are sorted in the second column and in the headline according to their size in descending order.

The median is between 9th and 10th value: [-0,49+(-0,50)]/2 = (-0,495). The numbers behind the fracture line represent the ranks.

The mean pairwise differences that not exceed the median are computed. From the table E.5 (EVS-EN 1500:2013) of critical values for Wilcoxon's matched-pairs signed-ranks test the entry for n=18 and for one-sided 0,025 level of significance, the critical value of 40 is found. Hence c = 40+1 = 41. The pairwise differences are sorted in descending order. The 41st value is -0.41. Hence the Hodges-Lehmann upper one-sided 97,5% confidence limit for the difference in lg Rs between RP and PP is -0.41, which is less than the agreed inferiority margin of 0,6. Therefore, the hypothesis of inferiority of PP is rejected and it can be concluded that the test preparation PP is non-inferior to RP.

Appendix 6

Interpretation

In the case of the reference substance the mean of the log of the pre-values (lg prevalues) is 5.93, which overrides the validation criterion ≥ 5. Tested product CHEMISEPT MED (Batch no. 196101017) shows the mean of the log of the pre-values (lg prevalues) 6.07, which is higher than the validation rate ≥ 5 as well.

There were not any of the lg reductions less than 3,00. The validation criterion is not more than three individual lg reductions less than 3,00 in RP.

Conclusion

Therefore, it can be claimed that by the validation criteria, test results and following counting: lg values of reduction, Wilcoxon test and Hodges-Lehmann system: the tested product – CHEMISEPT MED - is accepted for usage in hygienic handrub disinfection procedures on the following application: rub 3 ml of the product onto the hands within 30 seconds.

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