Inspection Report of VIRSHA (potassium hydrogen monopersulphate)

Sample name: VIRSHA (potassium hydrogen monopersulphate)
Sample quantity: 2 bottles; 250g/bottle
Requester: Hebei Erao Biotech Co.,Ltd
Sample properties: pink particles or powder
Producer: Hebei Erao Biotech Co.,Ltd
Sample collection date: 2023.01.07
Production date or batch number : 20221218
Testing completion date: 2023.05.27
Testing Lab: Qingdao Sci-tech Innovation Quality Testing Co.,Ltd

Testing Base

"Technical Specifications for Disinfection" (2002 Edition) 2.1.1.5.5
"Technical Specifications for Disinfection" (2002 Edition) 2.1.1.7
"Technical Specifications for Disinfection" (2002 Edition) 2.1.1.9
"Technical Specifications for Disinfection" (2002 Edition) 2.1.2.10

Evaluation basis

"Technical Specifications for Disinfection" (2002 Edition)

1 Test item: Stability test (composition)

1.1 Equipment

1.1.1 Test sample: VIRSHA , batch number: 20221218
1.1.2 Instruments and equipment: KC-SP-BL-001 stoppered buret, KC-SP-YQ-050 electronic balance.
1.1.3 Sodium thiosulfate standard solution, concentration is 0.1001 mol/L.

1.2 Method

1.2.1 Detection temperature is 20℃, relative humidity is 62%.

1.2.2 Test sample: VIRSHA, batch number: 20221218

1.3 Results

The average potassium hydrogen monopersulphate compound composition of VIRSHA is 54.28% (W/W). A batch of samples is measured, and each batch of samples is measured twice. The results are listed in Table 2.

Table 2 Determination results of potassium hydrogen monopersulphate compound composition

Sample serial number	potassium hydrogen monopersulphate compound composition (%, W/W)		Standard requirement
	Each time	Average	
1-1	54.111	54.28	50~60
1-2	54.444		

1.4 Conclusion

The average potassium hydrogen monopersulphate compound composition of the VIRSHA is 54.28% (W/W), which meets the standard requirements of the "Technical Specifications for Disinfection" .

2 Neutralizer identification test & bacterial suspension quantitative killing test

2.1 Equipment

2.1.1 Test sample: VIRSHA, batch number: 20221218

2.1.2 Test strains: Staphylococcus aureus (ATCC 6538) and Escherichia coli (8099), both of which were tested on their fifth-generation fresh slant cultures, provided by the Guangdong Provincial Food Microbial Safety Engineering Technology Research and Development Center.

2.1.3 Neutralizer: D/E neutralizing broth.

2.1.4 Organic interference: 3% (W/V) bovine serum albumin.

2.1.5 Instruments and equipment: KC-SP-YQ-013 biochemical incubator.

2.2 Method

2.2.1 Testing basis: "Technical Specifications for Disinfection" (2002 edition) 2.1.1.5 and 2.1.1.7.

2.2.2 Action concentration: diluent (active ingredient action concentration 1000mg/L).

2.2.3 Laboratory environment temperature: 20~22℃, humidity: 50~54%

2.2.4 Neutralizer identification test: The test bacterium is Escherichia coli (8099).

The test groups are:

(1) Disinfectant + bacterial suspension

(2) (disinfectant + bacterial suspension) + neutralizer

(3) Neutralizer + bacterial suspension
(4) (disinfectant + neutralizer) + bacterial solution
(5) Diluent + bacterial suspension
(6) Diluent from the same batch + neutralizer from the same batch + culture medium from the same batch.
VIRSHA, acting for 5 minutes, test temperature 19~21℃. The test was repeated three times.
2.2.5 Quantitative killing test: test bacteria Staphylococcus aureus (ATCC 6538), Escherichia coli (8099), action time is 5min, 10min, 15min, test temperature 19~21℃, test repeated 3 times, culture temperature 37.0℃.

2.3 Results

2.3.1. Bacterial neutralizer identification test

The average number of growing bacteria in group 1 is 0 CFU/mL, the average number of growing bacteria in group 2 was 5.00 × 102CFU/mL, and the error rates of colony numbers in groups 3, 4, and 5 were 2.70%, 1.30%, and 5.51% respectively. The serial numbers and contents of each group listed in the table are the same as the "Technical Specifications for Disinfection" (2002 edition). See Table 1 for details.

Table 1 E. coli neutralizer identification test results

Test serial number	Number of growing colonies in each group (CFU/mL)			Average number of growing colonies (CFU/mL)
	1	2	3	
1	0	0	0	0
2	2.70×10^2	4.90×10^2	7.40×10^2	5.00×10^2
3	1.49×10^7	2.75×10^7	4.10×10^7	2.78×10^7
4	1.42×10^7	2.69×10^7	3.70×10^7	2.60×10^7
5	1.53×10^7	2.79×10^7	4.30×10^7	2.87×10^7
6	0	0	0	0

Note: Negative control shows sterile growth.

2.3.2 Quantitative killing effect of bacteria

The test temperature was 19~21℃, and the test was repeated three times. The average killing log values of VIRSHA diluent (active ingredient concentration 1000mg/L) for 5 minutes on Staphylococcus aureus and Escherichia coli were 4.67/4.74 respectively, and the average killing log value for 10 minutes, the average killing log values of Staphylococcus aureus and Escherichia coli in 15 minutes are all >5.00. The data are listed in Table 2 and Table 3 respectively.

Table 2 Killing effect of sample dilution (active ingredient concentration 1000mg/L) on Staphylococcus aureus

Action time	Log killing value (KL) for different action times			Average log kill value (KL)
	1	2	3	

5min	4.67	4.68	4.67	4.67
10min	>5.00	>5.00	>5.00	>5.00
15min	>5.00	>5.00	>5.00	>5.00
positive control	7.11	7.32	7.53	7.32

Note: Negative control shows sterile growth.

Table 3 Killing effect of sample dilution (active ingredient concentration 1000 mg/L) on E. coli

Action time	Log killing value (KL) for different action times			Average log kill value (KL)
	1	2	3	
5min	4.75	4.73	4.74	4.74
10min	>5.00	>5.00	>5.00	>5.00
15min	>5.00	>5.00	>5.00	>5.00
positive control	7.25	7.39	7.66	7.43

Note: Negative control shows sterile growth.

2.4 Conclusion

2.4.1 The neutralizing agent identification test shows that: D/E neutralizing broth can effectively neutralize the bactericidal effect of VIRSHA diluent (active ingredient concentration 1000mg/L) on E. coli. The neutralizing agent and neutralization products are effective against E. coli. There are no adverse effects on the medium and culture medium, indicating that the neutralizer is suitable for quantitative killing tests of Escherichia coli and Staphylococcus aureus.

2.4.2 According to the "Technical Specifications for Disinfection" (2002 Edition), under test conditions, the average log killing value of VIRSHA diluent (active ingredient concentration 1000 mg/L) for 10 minutes against Staphylococcus aureus and Escherichia coli is > 5.00, in line with the standard requirements of "Technical Specifications for Disinfection" (2002 Edition).

3 Neutralizer identification test & Candida albicans suspension quantitative killing test

3.1 Equipment

3.1.1 Test sample: VIRSHA, batch number: 20221218

3.1.2 Test strain: Candida albicans (ATCC 10231) was tested on its third-generation fresh slant culture, which was provided by the Guangdong Provincial Food Microbial Safety Engineering Technology Research and Development Center.

3.1.3 Neutralizer: D/E neutralizing broth.

3.1.4 Organic interfering substances: 3% (W/V) bovine serum albumin.
3.1.5 Instruments and equipment: KC-SP-YQ-013 biochemical incubator.

3.2 Method

3.2.1 Testing basis: "Technical Specifications for Disinfection" (2002 edition) 2.1.1.5 and 2.1.1.9 .
3.2.2 Action concentration: diluent (active ingredient action concentration 1000mg/L).
3.2.3 Laboratory environment temperature: 20~22℃, humidity: 50~54%
3.2.4 Neutralizer identification test: The test bacteria is Candida albicans (ATCC 10231).
The test groups are:
(1) Disinfectant + bacterial suspension
(2) (disinfectant + bacterial suspension) + neutralizer
(3) Neutralizer + bacterial suspension
(4) (disinfectant + neutralizer) + bacterial solution
(5) Diluent + bacterial suspension
(6) Diluent from the same batch + neutralizer from the same batch + culture medium from the same batch.
VIRSHA, acting for 5 minutes, test temperature 19~21℃. The test was repeated three times.
3.2.5 Quantitative killing test: test bacteria Candida albicans (ATCC 10231), action time is 5min, 10min, 15min, test temperature 19~21℃, test repeated 3 times, culture temperature 37.0℃.

3.3 Results

3.3.1 Candida albicans neutralizing agent identification test
The average number of growing bacteria in group 1 was 0 CFU/mL, the average number of growing bacteria in group 2 was 5.60×10^2 CFU/mL, and the error rates of colony numbers in groups 3, 4, and 5 were 2.61%, 1.79%, and 7.88%. The serial numbers and contents of each group listed in the table are the same as the "Technical Specifications for Disinfection" (2002 edition). See Table 4 for details.

Table 4 Candida albicans neutralizer identification test results

Test serial number	Number of growing colonies in each group (CFU/mL)			Average growth colony Number (CFU/mL)
	1	2	3	
1	0	0	0	0
2	2.90×10^2	5.10×10^2	8.80×10^2	5.60×10^2
3	1.18×10^6	1.99×10^6	3.60×10^6	2.26×10^6
4	1.16×10^6	1.93×10^6	3.30×10^6	2.13×10^6
5	1.24×10^6	2.03×10^6	4.10×10^6	2.46×10^6
6	0	0	0	0

Note: Negative control shows sterile growth.

3.3.2. Quantitative killing effect of Candida albicans

The test temperature was 19~21℃, and the test was repeated three times. The average killing log value of VIRSHA diluent (active ingredient concentration 1000mg/L) for 5 minutes was 3.58, and the logarithmic killing value for Candida albicans was 3.58 for 10 minutes and 15 minutes. The average log killing values were all >4.00, and the data are listed in Table 5.

Table 5 sample diluent (active ingredient concentration 1000mg/L) killing effect on Candida albicans

Action time	Log killing value (KL) for different action times			Average log kill value (KL)
	1	2	3	
5min	3.58	3.59	3.57	3.58
10min	>4.00	>4.00	>4.00	>4.00
15min	>4.00	>4.00	>4.00	>4.00
positive control	6.25	6.09	6.39	6.24

Note: Negative control shows sterile growth

3.4 Conclusion

3.4.1 The neutralizing agent identification test shows that D/E neutralizing broth can effectively neutralize the bactericidal effect of VIRSHA diluent (active ingredient concentration 1000 mg/L) on Candida albicans. Candida and the culture medium have no adverse effects. It shows that the neutralizer is suitable for quantitative killing test of Candida albicans.

3.4.2 According to the "Technical Specifications for Disinfection" (2002 Edition), under test conditions, the average killing log value of VIRSHA diluent (active ingredient concentration 1000mg/L) for 10 minutes against Candida albicans is >4.00, which is in line with " "Technical Specifications for Disinfection" (2002 edition) standard requirements.

4 On-site identification test for disinfection of object surfaces by disinfectants (wooden surfaces)

4.1 Equipment

4.1.1 Test sample: VIRSHA

4.1.2 Neutralizer: D/E neutralizing broth.

4.1.3 Diluent: PBS solution containing 0.1% Tween 80.

4.1.4 Batch number: 20221218

4.1.5 Test equipment: sterile cotton swab, specification plate (5.0cm × 5.0cm).

4.1.6 Medium: Tryptone soy agar medium.

4.1.7 Instruments and equipment: KC-SP-YQ-013 biochemical incubator.

4.2 Method

4.2.1 Inspection basis: "Technical Specifications for Disinfection" (2002 edition) 2.1.2.10 .

4.2.2 Preparation of test solution: diluent (active ingredient concentration: 1000mg/L).

4.2.3 For each test, 30 samples of various items are tested on the surface. Randomly select the surface of the object (desktop, countertop, door, etc.), and use a specification board to mark two blocks with an area of 25 cm^2 each. One is for sampling before disinfection, and the other is for sampling after disinfection.

4.2.4 Before disinfection, moisten a sterile cotton swab in a test tube containing 5 mL of diluent, apply samples to an area, and go back and forth 8 times horizontally and vertically. After sampling, use aseptic operation to cut the sampling end of the cotton swab into the original diluent test tube, shake it for 20 seconds or beat it 80 times, and use it as a positive control sample after appropriate dilution.

4.2.5 According to the prescribed dosage, spray or apply the disinfectant on the surface of the object for disinfection. After disinfection, moisten the sterile cotton swab in a test tube containing 5 mL of neutralizer, apply samples to the disinfected area, and go back and forth 8 times horizontally and vertically. After sampling, cut the sampling end of the cotton swab into the test tube of the original sampling solution as a sample of the disinfection group.

4.2.6 Inoculate 1.0 mL each of the used neutralizer and diluent from the same batch into the culture medium as a negative control sample.

4.2.7 Inoculate 1.0 mL of each sample from the positive control group, negative control group, and disinfection group into 2 flat dishes, incubate at 37 ° C for 48 hours, and observe the results. Calculate the log kill value.

4.2.8 Test temperature is 21~22℃, relative humidity is 51~53%. The test was repeated three times.

4.3 Results

The test was repeated three times. Under the conditions of 21~22℃, the average logarithmic killing values of natural bacteria on the wooden surface were 1.32, 1.31 , and 1.31 respectively after wiping and disinfecting VIRSHA diluent (active ingredient concentration 1000 mg/L) for 10 minutes. List the data in Table 6, Table 7, and Table 8 respectively.

Table 6 On-site identification test results for wooden surface disinfection (Group 1)

Sample number	Number of colonies in the positive control group (CFU/cm^2)	Number of colonies in the test group (CFU/cm^2)	Log kill (KL)
1	21.20	1.10	1.28
2	8.60	0.70	1.09
3	7.00	0.50	1.15

4	16.10	0.80	1.30
5	8.70	0.50	1.24
6	6.50	0.20	1.51
7	16.20	1.00	1.21
8	25.90	0.70	1.57
9	8.30	0.50	1.22
10	7.90	0.50	1.20
11	23.00	1.10	1.32
12	14.70	0.50	1.47
13	16.80	1.00	1.23
14	23.90	1.00	1.38
15	16.00	0.70	1.36
16	16.00	0.50	1.51
17	12.00	0.50	1.38
18	23.00	1.10	1.32
19	7.40	0.50	1.17
20	7.70	0.30	1.41
21	25.90	0.90	1.46
22	10.70	0.50	1.33
23	15.70	0.90	1.24
24	4.90	0.30	1.21
25	25.60	0.90	1.45
26	17.60	0.70	1.40
27	20.20	1.00	1.31
28	22.40	1.20	1.27
29	7.80	0.50	1.19
30	26.10	0.90	1.46
Average Value	15.46	0.72	1.32
Note: The negative control group in each test showed sterile growth.			

Table 7 On-site identification test results for wooden surface disinfection (Group 2)

Sample number	Number of colonies in the positive control group (CFU/cm^2)	Number of colonies in the test group (CFU/cm^2)	Log kill (KL)
1	23.00	1.30	1.25
2	6.40	0.50	1.11
3	24.70	0.70	1.55
4	18.70	0.50	1.57

5	24.30	0.90	1.43
6	25.40	0.80	1.50
7	23.90	0.80	1.48
8	7.30	0.30	1.39
9	13.50	0.90	1.18
10	11.00	0.80	1.14
11	8.50	0.50	1.23
12	10.40	0.60	1.24
13	11.40	0.70	1.21
14	16.30	0.90	1.26
15	9.40	0.80	1.07
16	13.40	0.90	1.17
17	25.50	0.80	1.50
18	11.10	0.50	1.35
19	15.80	0.70	1.35
20	5.40	0.30	1.26
21	23.00	1.10	1.32
22	6.90	0.50	1.14
23	4.40	0.30	1.17
24	11.30	0.70	1.21
25	20.70	1.30	1.20
26	12.80	0.70	1.26
27	17.10	0.50	1.53
28	14.70	0.50	1.47
29	16.00	0.50	1.51
30	14.40	0.70	1.31
Average Value	14.89	0.70	1.31
Note: The negative control group in each test showed sterile growth.			

Table 8 On-site identification test results for wooden surface disinfection (Group 3)

Sample number	Number of colonies in the positive control group (CFU/cm^2)	Number of colonies in the test group (CFU/cm^2)	Log kill (KL)
1	15.50	0.70	1.35
2	8.40	0.50	1.23
3	6.10	0.30	1.31
4	20.00	0.90	1.35

5	15.40	0.80	1.28
6	11.30	0.50	1.35
7	24.80	1.00	1.39
8	11.80	0.70	1.23
9	25.30	1.40	1.26
10	11.50	0.70	1.22
11	12.90	0.50	1.41
12	6.70	0.30	1.35
13	4.20	0.20	1.32
14	20.60	0.90	1.36
15	22.50	1.00	1.35
16	16.20	0.70	1.36
17	7.40	0.50	1.17
18	4.80	0.30	1.20
19	8.40	0.70	1.08
20	24.70	1.10	1.35
21	22.50	0.90	1.40
22	14.40	0.50	1.46
23	24.20	0.80	1.48
24	7.70	0.50	1.19
25	18.70	0.60	1.49
26	17.40	1.00	1.24
27	21.40	1.20	1.25
28	20.50	1.60	1.11
29	12.60	0.50	1.40
30	10.50	0.60	1.24
Average Value	14.95	0.73	1.31
Note: The negative control group in each test showed sterile growth.			

4.4. Conclusion

The field test of wooden surface disinfection shows that under the conditions of 21~22℃, the average killing log value of natural bacteria on the wooden surface after 10 minutes of wipe disinfection with VIRSHA diluent (active ingredient concentration 1000mg/L) is >1.00, which is in line with the " "Technical Specifications for Disinfection" (2002 edition) standard requirements.

Authorized technical person in charge (signature): 陈雷

Final review date: MAY 27, 2023 Stamp for inspection and testing: