CMA
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Inspect report

Sample No.:2020-XD-217
Sample name: VIRSHA (potassium monopersulphate)
Sample quantity: 8 bottles; 250g/bottle
Requester: Hebei Erao Biotech Co.,Ltd
Sample properties: pink particles or powder
Producer: Hebei Erao Biotech Co.,Ltd
Production date or batch number: 20200208
Test lab: Zhongguancun International Medical Inspection and Certification Technology Co., Ltd.

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Metal corrosion determination

Sample name: VIRSHA(potassium monopersulphate)
Inspection item: Metal corrosion determination
Sample acceptance date: February 24, 2020
Inspection completion date: November 20, 2020

1. Equipment
1.1 VIRSHA (potassium monopersulphate) (powder; active ingredients: available chlorine, available oxygen, potassium monopersulfate compound; production date/batch number: 20200208).
1.2. Electronic balance (No. LH029), electric constant temperature drying oven (No. LH032).
1.3. Metal sheet: round, 24±0.1mm in diameter, 1.0mm thick, pierced with a 2.0mm diameter hole, with a total surface area of about 9.8cm², and a smoothness of 4 to 6; the raw materials are as follows: carbon steel (GB700 -65), copper (GB2060-80), aluminum (GB1173-74), stainless steel (GB1220-75).
1.4. Wide-mouth reagent bottle (1000mL), volumetric flask (100mL), sandpaper (No. 120 grit water sandpaper, GB2477).
1.5. Detergent, absolute ethanol.

2. Method
2.1. Test basis: Item 2.2.4 of "Technical Specifications for Disinfection" (2002 Edition).
2.2. Test method: Soak the metal piece in detergent for 10 minutes to fully remove oil and wash it. Use sandpaper to remove the oxide layer on the surface of the metal sheet and rinse with tap water. Degrease with absolute ethanol, dry in a 50°C incubator for 1 hour, and weigh. Tie the metal piece with a plastic thread to label, number and date it, hang it and soak it in a wide-mouth reagent bottle containing the sample solution (prepare the sample with water into a sample solution with an effective chlorine content of 4000 mg/L), and soak 72h. After soaking for the specified time, take out the metal piece, rinse it with tap water, and use a brush to remove the corrosion products and clean it. After the coarse filter paper absorbs the water, place it in a 50°C incubator to dry for 1 hour and weigh. The test also included a blank control of stainless steel soaked in distilled water. Each 600mL sample.
This product uses liquid to soak 3 pieces of the same metal piece, with each piece separated by more than 1cm. Each piece of sandpaper only sands one metal material.
2.3. Detection ambient temperature: 23.5℃, relative humidity: 46.0%.

3. Results
Under the conditions of ambient temperature of 23.5℃ and relative humidity of 46.0%, metal corrosion testing was performed on VIRSHA (potassium monopersulphate) (available chlorine content is 4000mg/L). The corrosion rate of this sample to stainless steel metal sheets is 0.0000mm/ a; The corrosion rate for carbon steel metal sheets is 4.7805mm/a; the corrosion rate for copper metal sheets is 7.8343mm/a; the corrosion rate for aluminum metal sheets is 8.5755mm/a (see attached table).

Scheduled metal corrosion test results

Metal type	Sample measurement average			Corrosion rate R(mm/a)	Corrosivity level
	Weigh the sample before testing (g/piece)	Weigh the sample after the test (g/piece)	Weight loss value (g/piece)		
Stainless steel	3.1562	3.1562	0.0000	0.0000	Basically no corrosion
Carbon steel	3.3650	3.0635	0.3015	4.7805	Severe corrosion
Copper	3.6523	3.1115	0.5408	7.8343	Severe corrosion
Lead	1.1314	0.9449	0.1865	8.5755	Severe corrosion

4. Conclusion
After testing, VIRSHA (potassium monopersulphate) (available chlorine content is 4000mg/L) is basically non-corrosive to stainless steel, but severely corrosive to carbon steel, copper and aluminum.

Pseudomonas aeruginosa suspension quantitative killing test

Sample name: VIRSHA(potassium monopersulphate)
Test item: Pseudomonas aeruginosa suspension quantitative killing test
Sample acceptance date: February 24, 2020
Inspection completion date: March 27, 2020

1. Equipment
1.1. Test strain: Pseudomonas aeruginosa (ATCC15442), 8th generation.
1.2. VIRSHA (potassium monopersulphate) (powder; active ingredients: available chlorine, available oxygen , potassium monopersulfate compound; production date/batch number: 20200208).
1.3. Neutralizer composition and concentration: PBS containing 5g/L sodium thiosulfate.
1.4. Diluent: tryptone saline solution (TPS).
1.5. Organic interfering substances: 3% bovine serum albumin (BSA).
1.6. Tryptone soy agar (TSA).
1.7. Biological safety cabinets, constant temperature incubators, thermostats, vortex oscillators, sterile equipment and electronic timers, etc.

2. Method
2.1. Testing basis: Item 2.1.1.7.4 of "Technical Specifications for Disinfection" (2002 Edition).
2.2. Sterilization test: Use standard hard water to dissolve VIRSHA (potassium monopersulphate) and dilute it into a test solution with an effective chlorine content of 375mg/L (1.25 times the concentration to be measured, the actual concentration is 300mg/L), and the action time are 5.0min, 10.0min and 15.0min, and the test temperature is a constant temperature of 20℃. The test was repeated three times.
2.3. Detection of ambient temperature: 20.5℃~20.9℃; relative humidity: 47%.

3. Results
After three repeated tests, under the constant temperature test conditions of 20°C, VIRSHA (potassium monopersulphate) test solution with an effective chlorine content of 300 mg/L was applied for 10.0 minutes to eliminate the effects of Pseudomonas aeruginosa in the suspension. The logarithmic killing values are all >5.00 (see attached table).

Attached table of killing effects against Pseudomonas aeruginosa

Experiment serial number	Value for different time periods (min)			Number of positive control colonies
	5.0	10.0	15.0	(cfu/mL)
1	>5.00	>5.00	>5.00	1.67×10^7
2	>5.00	>5.00	>5.00	1.54×10^7
3	>5.00	>5.00	>5.00	1.27×10^7

Note: Negative control shows sterile growth.

4. Conclusion
After three repeated tests, under the constant temperature test conditions of 20°C, VIRSHA (potassium monopersulphate) test solution with an effective chlorine content of 300 mg/L was applied for 10.0 minutes to eliminate the effects of Pseudomonas aeruginosa in the suspension. The log killing values were all >5.00. It complies with the disinfection qualification requirements of the "Technical Specifications for Disinfection" (2002 Edition).

Quantitative killing test of Bacillus subtilis black variant spore suspension

Sample name: VIRSHA(potassium monopersulphate)
Test items: Quantitative killing test of Bacillus subtilis black variant spore suspension
Sample acceptance date: February 24, 2020
Inspection completion date: March 27, 2020

1. Equipment
1.1. Test strain: Bacillus subtilis var black (ATCC9372) spores.
1.2. VIRSHA (potassium monopersulphate) (powder; active ingredients: available chlorine, available oxygen, potassium monopersulfate compound; production date/batch number: 20200208).
1.3. Neutralizer composition and concentration: PBS containing 5g/L sodium thiosulfate.
1.4. Diluent: tryptone saline solution (TPS).
1.5. Organic interfering substances: 3% bovine serum albumin (BSA).
1.6. Tryptone soy agar (TSA).
1.7. Biological safety cabinets, incubators, thermostats, vortex oscillators, sterile equipment and electronic timers, etc.

2. Method
2.1. Testing basis: Item 2.1.1.5.5 and Item 2.1.1.7.4 of "Technical Specifications for Disinfection" (2002 Edition).
2.2. Neutralizer identification test
Use standard hard water to dissolve VIRSHA (potassium monopersulphate) and dilute it into a test solution with an effective chlorine content of 5000mg/L (1.25 times the concentration to be measured, the actual concentration is 4000mg/L). The action time is 60.0min. Test temperature is constant at 20°C. The test was repeated three times.
2.3. Sterilization test
Use standard hard water to dissolve VIRSHA (potassium monopersulphate) and dilute it into a test solution with an effective chlorine content of 5000mg/L (1.25 times the concentration to be measured, the actual concentration is 4000mg/L), and the action time is 60.0min, 120.0 min and 180.0min, the test temperature is a constant temperature of 20℃. The test was repeated three times.
2.4. Detection of ambient temperature: 20.5℃~22.3℃; relative humidity: 46%~47%.

3. Results
3.1. Neutralizer identification test
After three repeated tests, under the constant temperature test conditions of 20°C, the neutralizer identification result of the VIRSHA (potassium monopersulphate) test solution with an effective chlorine content of 4000 mg/L was : the average number of colonies in the first group was 0cfu/mL, the average number of colonies in group 2 was 2.28×10^4 cfu/mL, and the average number of colonies in groups 3, 4 and 5 were 1.34×10^7cfu/mL, 1.24×10^7cfu/mL and 1.40×10^7cfu/mL respectively, the error rate among the three groups was 4.38 % , and the 6th group was the negative control.

Schedule of neutralizer identification test results

Group	The growth rate (cfumL) of the three test groups			Average value (cfu/mL)	Between groups 3, 4, and 5 Error rate(%)
	1	2	3		
1	0	0	0	0	4.38
2	2.24×10^4	2.11×10^4	2.50×10^4	2.28×10^4	
3	1.34×10^7	1.47×10^7	1.22×10^7	1.34×10^7	
4	1.23×10^7	1.37×10^7	1.13×10^7	1.24×10^7	
5	1.37×10^7	1.53×10^7	1.31×10^7	1.40×10^7	
6	0	0	0	0	

3.2. Killing effect on Bacillus subtilis black variant spores
After three repeated tests, under the constant temperature test conditions of 20°C, the test solution of VIRSHA (potassium monopersulphate) with an effective chlorine content of 4000 mg/L was applied , and the action time was 120.0 min. The log killing values of spores of the Bacillus subtilis black variant in the suspension were all >5.00.

Attached table: Killing effect on Bacillus subtilis var. black spores

Serial number	Killing value for different time (min)			Number of positive control colonies(cfu/mL)
	60.0	120.0	180.0	
1	2.88	>5.00	>5.00	1.66×10^7
2	2.87	>5.00	>5.00	1.77×10^7
3	2.85	>5.00	>5.00	1.62×10^7

Note: Negative control shows sterile growth.

4. Conclusion

4.1. After three repeated tests, under the constant temperature test conditions of 20°C, the neutralizer solution containing 5g/L sodium thiosulfate in PBS can effectively neutralize VIRSHA (potassium monopersulphate) with an effective chlorine content of 4000mg/L. The test solution of VIRSHA (potassium monopersulphate) has a residual effect on the spores of Bacillus subtilis var. black, and the neutralizer and its neutralization products have no adverse effects on the culture medium and have basically no effect on the growth of spores of Bacillus subtilis var. black.

4.2. After three repeated tests, under the constant temperature test conditions of 20°C, apply the test solution of VIRSHA (potassium monopersulphate) with an effective chlorine content of 4000 mg/L for 120.0 minutes. The log killing values of B. subtilis var. black spores in the suspension were all >5.00. It complies with the disinfection qualification requirements of the "Technical Specifications for Disinfection" (2002 Edition).

Quantitative inactivation test of poliovirus suspension

Sample name: VIRSHA(potassium monopersulphate)
Test items: Quantitative inactivation test of poliovirus suspension
Sample acceptance date: February 24, 2020
Inspection completion date: December 28 , 2020

1. Equipment
1.1. Test virus strain: poliovirus type I (poliovirus-I, PV-I) vaccine strain.
1.2. Host cell: VERO cells.
1.3. VIRSHA (potassium monopersulphate) (powder; active ingredients: available chlorine, available oxygen, potassium monopersulfate compound; production date/batch number: 20200208).
1.4. Neutralizer composition and concentration: PBS containing 5g/L sodium thiosulfate.
1.5. Cell culture flask and 96-well culture plate.
1.6. Organic interfering substances: 3% bovine serum albumin (BSA).
1.7. Cell maintenance culture medium, complete cell culture medium and fetal bovine serum.
1.8. Thermostat, inverted microscope, carbon dioxide incubator, biological safety cabinet, micropipette and sterile equipment.

2. Method
2.1. Testing basis : Item 2.1.1.10.5 and Item 2.1.1.10.7 of "Technical Specifications for Disinfection" (2002 Edition).
2.2. Preparation of virus suspension: dilute the PV-I virus suspension with a titer of 10^6TCID_{50}/0.1mL to 10^7TCID_{50}/0.1mL and 3% bovine serum albumin organic interfering substance, and set it at a constant temperature of 20°C for later use.
2.3. Neutralizer identification test: Use standard hard water to dissolve VIRSHA (potassium monopersulphate) and dilute it into a test solution with an effective chlorine content of 1250mg/L (1.25 times the concentration to be measured, the actual concentration is 1000mg/L), the action time is 10.0min, and the test temperature is a constant temperature of 20°C. The test was repeated three times.
2.4. Virus inactivation test: Use standard hard water to dissolve VIRSHA (potassium monopersulphate) and dilute it into a test solution with an effective chlorine content of 1250mg/L (1.25 times the concentration to be measured, the actual concentration is 1000mg/L), the action time is 10.0min, 20.0min and 30.0min, and the test temperature is a constant temperature of 20℃. The test was repeated three times.
2.5. Detection of ambient temperature: 25.0℃~25.3℃; relative humidity: 45%~46%.

3. Results
3.1. Neutralizer identification test
After three repeated tests, under the constant temperature test conditions of 20°C, the test liquid neutralizer identification result of VIRSHA (potassium monopersulphate) with an effective chlorine content of 1000 mg/L was: the average titer value of the virus in Group 1 was 2.44, the average virus titer value in group 2 was 2.61, the average virus titer value in groups 3, 4 and 5 were 6.72, 6.56 and 6.61 respectively, and group 6 was the negative control group.

Schedule of neutralizer identification test results

Group	Virus titer value ($TCID_{50}$) in each group of three tests			Average virus titer value($TCID_{50}$)
	1	2	3	
1	2.5	2.5	2.33	2.44
2	2.67	2.67	2.5	2.61
3	6.67	7	6.5	6.72
4	6.5	6.67	6.5	6.56
5	6.67	6.67	6.5	6.61
6	0	0	0	0

3.2. Inactivation effect on poliovirus
After three repeated tests, under the constant temperature test conditions of 20°C, the test solution of VIRSHA (potassium monopersulphate) with an effective chlorine content of 1000 mg/L was applied for 20.0 minutes, and the log value of the inactivation of poliovirus was obtained. All >4.00.

Schedule of inactivating efficacy against poliovirus type I vaccine strains

Serial number	For different time (min)	Positive control

	10.0	20.0	30.0	virus titer value ($TCID_{50}$)
1	4.00	>4.00	>4.00	6.67
2	4.00	>4.00	>4.00	6.67
3	4.00	>4.00	>4.00	6.50

Note: Negative control cells grow well and have no cell lesions.

4. Conclusion

4.1. After three repeated tests, under the constant temperature test conditions of 20°C, the neutralizer solution used in PBS containing 5g/L sodium thiosulfate can effectively neutralize VIRSHA (potassium hydrogen) with an effective chlorine content of 1000mg/L. monopersulphate) has a residual effect on the poliovirus type I vaccine strain, and the neutralizer and neutralization products have basically no effect on the growth of the poliovirus type I vaccine strain and cells.

4.2. After three repeated tests, under the constant temperature test conditions of 20°C, VIRSHA (potassium monopersulphate) test solution with an effective chlorine content of 1000 mg/L was applied for 20.0 minutes, and the effect on poliovirus type I vaccine was The inactivation logarithm values of the strains were all >4.00, which complied with the requirements for qualified disinfection in the "Technical Specifications for Disinfection" (2002 Edition).

On-site test of surface disinfection of food processing equipment

Sample name: VIRSHA(potassium monopersulphate)
Inspection item: On-site test of surface disinfection of food processing equipment
Sample acceptance date: February 24, 2020
Inspection completion date: April 24, 2020

1. Equipment
1.1. Test strains: natural bacteria on the surface of objects.
1.2. VIRSHA (potassium monopersulphate) (powder; active ingredients: available chlorine, available oxygen, potassium monopersulfate compound; production date/batch number: 20200208).
1.3. Neutralizer composition and concentration: PBS containing 5g/L sodium thiosulfate.
1.4. Diluent: 0.03mol/L phosphate buffer containing 0.1% Tween 80.
1.5. Tryptone soy agar (TSA).
1.6. Specification board (5.0cm×5.0cm).
1.7. Object surface: surface of food processing equipment.
1.8. Biological safety cabinets, incubators, vortex oscillators, sterile equipment and electronic timers, etc.

2. Method
2.1. Test basis: Item 2.1.2.10 of "Technical Specifications for Disinfection" (2002 Edition).
2.2. Test method: Randomly select the surface of the food processing equipment, use a specification board to mark two adjacent areas with an area of 25cm², use sterile cotton swabs to sample and prepare sample liquid before disinfection, as a control group. Dip sterile gauze into VIRSHA (potassium monopersulphate) test solution with an effective chlorine content of 300mg/L, scrub another area repeatedly, disinfect for 15.0 minutes, use a sterile cotton swab containing neutralizer to take samples and Prepare a sample solution as a test group. Use the same batch of neutralizer and diluent as the negative control group. After the sample liquids of the control group and the test group were fully shaken, 1.0 mL inoculation plates were drawn into the plates in duplicate, and TSA agar was poured into them. After condensation, they were placed in a 37°C incubator and cultured for 48 hours, and the number of viable bacteria was counted.
2.3. Detection ambient temperature: 22.0℃, relative humidity: 46%.

3. Results
Under the conditions of ambient temperature of 22.0°C and relative humidity of 46%, VIRSHA (potassium monopersulphate) test fluid with an effective chlorine content of 300 mg/L was applied for 15.0 minutes to naturally affect the surface of 30 food processing equipment samples. The average killing log value of bacteria is 1.31, ranging from 0.94 to 1.89 (see attached table).

Attached table: Field test results for surface disinfection of objects

Series number	The number of colonies in the control group (cfu/25cm²)	Number of colonies in the test group (cfu/25cm²)	Log kill value
1	6.4×10³	2.8×10²	1.36
2	5.95×10³	6.05×10²	0.99
3	6.2×10³	1.6×10²	1.59
4	6.3×10³	6.05×10²	1.02
5	1.09×10³	1.4×10²	1.89
6	1.13×10³	5.6×10²	1.3
7	4.65×10³	2.6×10²	1.25
8	7.95×10³	4.15×10²	1.28
9	6.85×10³	3.55×10²	1.29
10	5.85×10³	6.00×10²	0.99
11	9.1×10³	6.1×10²	1.17
12	6.3×10³	1.85×10²	1.53
13	9.55×10³	5.85×10²	1.21

14	6.3×10^3	1.15×10^2	1.74
15	5.65×10^3	3.55×10^2	1.2
16	7.15×10^3	4.15×10^2	1.23
17	5.8×10^3	2.85×10^2	1.31
18	9.4×10^3	1.85×10^2	1.7
19	8.85×10^3	3.25×10^2	1.44
20	2.0×10^3	2.0×10^2	1.00
21	7.65×10^3	4.0×10^2	1.28
22	3.55×10^3	2.4×10^2	1.17
23	6.05×10^3	5.45×10^2	1.04
24	7.4×10^3	2.7×10^2	1.44
25	5.85×10^3	3.15×10^2	1.27
26	4.3×10^3	4.9×10^2	0.94
27	4.8×10^3	3.35×10^2	1.16
28	5.45×10^3	2.0×10^2	1.44
29	7.1×10^3	2.5×10^2	1.45
30	3.2×10^3	1.1×10^2	1.47
Average			1.31

4. Conclusion

After testing, the test solution of VIRSHA (potassium monopersulphate) with an effective chlorine content of 300mg/L, which was used for 15.0 minutes, had an average killing log value of >1.00 of natural bacteria on 30 food processing equipment surface samples, which complies with the "Technical Specifications for Disinfection" (2002 edition).

Food (drinking) utensils disinfection simulation field test

Sample name: VIRSHA (potassium monopersulphate)
Inspection items: Food (drinking) utensils disinfection simulation field test
Sample acceptance date: February 24, 2020
Inspection completion date: May 9, 2020

1. Equipment
1.1. Test strain: Escherichia coli (8099), 8th generation.
1.2. VIRSHA (potassium monopersulphate) (powder; active ingredients: available chlorine, available oxygen, potassium monopersulfate compound; production date/batch number: 20200208).
1.3. Neutralizer composition and concentration: PBS containing 5g/L sodium thiosulfate.
1.4. Diluent: phosphate buffer saline (PBS, 0.03M, pH7.2).
1.5. Tryptone soy agar (TSA).
1.6. Specification board (5.0cm×5.0cm), 6L plastic container.
1.7. Food (drink) utensils: porcelain bowl (after pressure steam sterilization, dry and set aside).
1.8. Biological safety cabinets, incubators, vortex oscillators, sterile equipment and electronic timers, etc.

2. Method
2.1. Testing basis: Item 2.1.2.1 of "Technical Specifications for Disinfection" (2002 Edition).
2.2. Test method: Use a specification board to mark the bacteria-contaminated area (5.0cm×5.0cm) on the inside of the porcelain bowl for testing, add 0.1mL of bacterial solution, spread evenly with a sterile L rod, and perform the test after drying. Immerse 30 bacteria-contaminated porcelain bowls in 6L plastic containers filled with VIRSHA (potassium monopersulphate) test solution with an effective chlorine content of 1000mg/L, soak and disinfect for 20.0 minutes, take them out, and add them to the porcelain bowls. Add 5 mL of neutralizer solution to the infected area and pipe thoroughly for 10.0 minutes to serve as the disinfection test group. The three bacteria-contaminated porcelain bowls were not soaked in disinfectant, and 5 mL of neutralizer solution was directly added to the bacteria-contaminated area in the porcelain bowls. After the disinfection treatment of the test group was completed, sampling and testing were conducted along with the test group as a positive control group. Aspirate the neutralizer and diluent used in the same test to inoculate the plate as a negative control group. Take 1.0 mL of each of the above groups and inoculate the plates in duplicate. After inoculating the plates, pour TSA agar into each group. After condensation, place it in a 37°C incubator and culture it for 48 hours, and count the number of viable bacteria.
2.3. Detection ambient temperature: 22.0℃, relative humidity: 46%.

3. Results
Under the conditions of ambient temperature of 22.0°C and relative humidity of 46%, apply VIRSHA (potassium monopersulphate) test solution with an effective chlorine content of 1000 mg/L, soak and disinfect for 20.0 minutes, and test the large intestine on 30 porcelain bowl surface samples. The average log killing value of bacilli is 7.46 (see attached table).

Attached table: Simulation field test results for disinfection of food (drinking) utensils

Sample number	The number of colonies in the test group(cfu/25cm²)	The number of colonies in the control group(cfu/25cm²)	Log kill value
1	0	2.88×10^7 (2.70×10^7~3.05×10^7)	7.46
2	0		7.46
3	0		7.46
4	0		7.46
5	0		7.46
6	0		7.46
7	0		7.46
8	0		7.46
9	0		7.46
10	0		7.46
11	0		7.46

12	0		7.46
13	0		7.46
14	0		7.46
15	0		7.46
16	0		7.46
17	0		7.46
18	0		7.46
19	0		7.46
20	0		7.46
21	0		7.46
22	0		7.46
23	0		7.46
24	0		7.46
25	0		7.46
26	0		7.46
27	0		7.46
28	0		7.46
29	0		7.46
30	0		7.46
Average			7.46

Note: Negative control shows sterile growth.

4. Conclusion

After testing, the test solution of VIRSHA (potassium monopersulphate) with an effective chlorine content of 1000mg/L was used, soaked and disinfected for 20.0 minutes, and the logarithmic killing value of E. coli on 30 porcelain bowl surface samples was all >3.00, in line with the " "Technical Specifications for Disinfection" (2002 edition).

Fabric surface disinfection simulation field test

Sample name: VIRSHA (potassium monopersulphate)
Inspection item: Fabric surface disinfection simulation field test
Sample acceptance date: February 24, 2020
Inspection completion date: May 14, 2020

1. Equipment
1.1. Test strain: Escherichia coli (8099), 8th generation.
1.2. VIRSHA (potassium monopersulphate) (powder; active ingredients: available chlorine, available oxygen, potassium monopersulfate compound; production date/batch number: 20200208).
1.3. Neutralizer composition and concentration: PBS containing 5g/L sodium thiosulfate.
1.4. Diluent: 0.03mol/L phosphate buffer containing 0.1% Tween 80.
1.5. Tryptone soy agar (TSA).
1.6. Specification board (5.0cm×5.0cm).
1.7. Fabric surface: White coat fabric surface.
1.8. Biological safety cabinets, incubators, vortex oscillators, sterile equipment and electronic timers, etc.

2. Method
2.1. Inspection basis: Item 2.1.2.9 of "Technical Specifications for Disinfection" (2002 Edition).
2.2. Test method: Randomly select the surface of the white coat fabric, use a specification board to mark 60 bacteria-contaminated areas with an area of 25cm², apply the bacterial suspension evenly to the bacteria-contaminated areas, and wait for natural drying before conducting the test. Before disinfection, use sterile cotton swabs to collect 30 infected areas and prepare sample solutions as a positive control group; soak the infected clothes in VIRSHA (potassium monopersulphate) with an effective chlorine content of 300mg/L. In the solution, disinfect for 10.0 minutes, take samples with sterile cotton swabs containing neutralizer and prepare sample solutions as the test group. The neutralizer, diluent and cotton swab eluate used in the same batch of tests were used as negative control groups. After the sample liquids of the control group and the test group were fully shaken, 1.0 mL inoculation plates were drawn into the plates in duplicate, and TSA agar was poured into them. After condensation, they were placed in a 37°C incubator and cultured for 48 hours, and the number of viable bacteria was counted.
2.3. Detection of ambient temperature: 22.1℃; relative humidity: 46%.

3. Results
Under the conditions of ambient temperature of 22.1°C and relative humidity of 46%, apply VIRSHA (potassium monopersulphate) test solution with an effective chlorine content of 300 mg/L, soak and disinfect for 10.0 minutes, and test the surface of 30 white coat fabric samples. The average killing log value of E. coli is 7.11, with a range of 5.95~7.39 (see attached table).

Attached table: Simulation field test results for fabric surface disinfection

Sample number	The number of colonies in the test group(cfu/25cm²)	The number of colonies in the control group(cfu/25cm²)	Log kill value
1	1.35×10^7	0	7.13
2	1.46×10^7	0	7.16
3	1.29×10^7	0	7.11
4	1.33×10^7	0	7.12
5	1.36×10^7	15	5.95
6	1.90×10^7	5	6.58
7	2.05×10^7	0	7.31
8	2.15×10^7	5	6.63
9	1.90×10^7	0	7.28
10	1.85×10^7	0	7.27
11	2.10×10^7	0	7.32
12	1.65×10^7	0	7.22
13	1.60×10^7	0	7.2

14	1.85×10^7	0	7.27
15	1.95×10^7	0	7.29
16	2.20×10^7	0	7.34
17	2.00×10^7	0	7.3
18	1.95×10^7	15	6.11
19	2.35×10^7	0	7.37
20	1.60×10^7	0	7.2
21	1.85×10^7	0	7.27
22	2.10×10^7	0	7.32
23	2.25×10^7	0	7.35
24	2.35×10^7	0	7.37
25	2.25×10^7	0	7.35
26	2.10×10^7	15	6.14
27	2.45×10^7	0	7.39
28	2.40×10^7	0	7.38
29	2.35×10^7	0	7.37
30	2.00×10^7	0	7.3
Average			7.11

Note: Negative control shows sterile growth.

4. Conclusion

After testing, the test solution of VIRSHA (potassium monopersulphate) with an effective chlorine content of 300mg/L was used, soaked and disinfected for 10.0 minutes, and the log kill value of E. coli on 30 white coat fabric surface samples was all >3.00. It complies with the disinfection qualification requirements of the "Technical Specifications for Disinfection" (2002 Edition).

On-site test on surface disinfection of fruits and vegetables

Sample name: VIRSHA (potassium monopersulphate)
Inspection items: On-site test on surface disinfection of fruits and vegetables
Sample acceptance date: February 24, 2020
Inspection completion date: May 9, 2020

1. Equipment
1.1. Test strains: natural bacteria on the surface of fruits and vegetables.
1.2. VIRSHA (potassium monopersulphate) (powder; active ingredients: available chlorine, available oxygen, potassium monopersulfate compound; production date/batch number: 20200208).
1.3. Neutralizer composition and concentration: PBS containing 5g/L sodium thiosulfate.
1.4. Diluent: 0.03mol/L phosphate buffer containing 0.1% Tween 80.
1.5. Tryptone soy agar (TSA).
1.6. Specification board (5.0cm×5.0cm).
1.7. Surface: Dutch cucumber.
1.8. Biological safety cabinets, incubators, vortex oscillators, sterile equipment and electronic timers, etc.

2. Method
1. Test basis: Item 2.1.2.10 of "Technical Specifications for Disinfection" (2002 Edition).
2. Test method: Randomly select Dutch cucumbers, use a specification board to mark two adjacent areas with an area of 25cm², use sterile cotton swabs to sample and prepare a sample solution before disinfection, as a control group. Soak another area in VIRSHA (potassium monopersulphate) test solution with an effective chlorine content of 300mg/L, disinfect it for 15.0 minutes, use a sterile cotton swab containing neutralizer to sample and prepare a sample solution, as test group. Use the same batch of neutralizer, diluent, and cotton swab eluate as negative control groups. After the sample liquids of the control group and the test group were fully shaken, 1.0 mL inoculation plates were drawn into the plates in duplicate, and TSA agar was poured into them. After condensation, they were placed in a 37°C incubator and cultured for 48 hours, and the number of viable bacteria was counted.
3. Detection ambient temperature: 22.0℃, relative humidity: 46%.

3. Results
Under the conditions of ambient temperature of 22.0°C and relative humidity of 46%, VIRSHA (potassium monopersulphate) test solution with an effective chlorine content of 300mg/L was applied for 15.0 minutes to control the natural bacteria on the surface of 30 Dutch cucumber samples. The average log killing value is 1.60, with a range of 0.80~2.19 (see attached table).

Attached table: field test results for surface disinfection of fruits and vegetables

Sample number	The number of colonies in the test group(cfu/25cm²)	The number of colonies in the control group(cfu/25cm²)	Log kill value
1	1.28×10^{6}	3.40×10^{4}	1.58
2	1.03×10^{6}	2.00×10^{4}	1.71
3	1.65×10^{5}	2.60×10^{4}	0.80
4	5.35×10^{5}	1.05×10^{4}	1.71
5	1.09×10^{6}	1.19×10^{4}	1.96
6	1.08×10^{6}	3.45×10^{4}	1.49
7	4.60×10^{5}	1.65×10^{4}	1.44
8	7.55×10^{5}	2.00×10^{4}	1.58
9	1.16×10^{6}	3.55×10^{4}	1.51
10	6.15×10^{5}	2.45×10^{4}	1.40
11	8.85×10^{5}	2.85×10^{4}	1.50
12	1.13×10^{6}	1.90×10^{4}	1.77
13	8.95×10^{5}	3.10×10^{4}	1.46
14	5.65×10^{5}	1.65×10^{4}	1.53
15	1.06×10^{6}	3.55×10^{4}	1.48
16	4.15×10^{5}	4.15×10^{4}	1.00
17	1.80×10^{5}	6.15×10^{3}	1.47
18	9.90×10^{5}	7.0×10^{3}	2.15
19	5.05×10^{5}	7.70×10^{3}	1.81
20	7.25×10^{5}	1.04×10^{3}	1.84

21	1.26×10^5	4.05×10^3	1.49
22	1.08×10^5	2.30×10	1.67
23	6.30×10^4	5.20×10^3	1.08
24	4.15×10^5	2.70×10^3	2.19
25	3.70×10^5	3.15×10^3	2.07
26	2.80×10^5	4.81×10^3	1.76
27	3.30×10^5	1.60×10^4	1.32
28	1.03×10^6	2.00×10^4	1.71
29	8.80×10^5	2.50×10^4	1.54
30	3.60×10^5	4.8×10^4	1.88
Average			1.60

Note: Negative control shows sterile growth.

4. Conclusion

After testing, the test solution of VIRSHA (potassium monopersulphate) with an effective chlorine content of 300mg/L was used for 15.0 minutes. The average killing log value of natural bacteria on 30 Dutch cucumber surface samples was >1.00, which complies with the "Technical Specifications for Disinfection" (2002 edition).

Air disinfection simulation field test

Sample name: VIRSHA (potassium monopersulphate)
Inspection items: Air disinfection simulation field test
Sample acceptance date: February 24, 2020
Inspection completion date: June 17, 2020

1. Equipment
1.1. Test strain: Staphylococcus albus (8032), 7th generation.
1.2. VIRSHA (potassium monopersulphate) (powder; active ingredients: available chlorine, available oxygen, potassium monopersulfate compound; production date/batch number: 20200208).
1.3.Aerosol chamber volume: 20m³.
1.4. Sampling device: six-stage sieve air impact sampler.
1.5. Culture medium: ordinary nutrient broth medium, ordinary nutrient agar medium, ordinary nutrient agar medium containing neutralizer (neutralizer ingredient: D/E neutralizing broth).
1.6. Biological safety cabinets, incubators, vortex oscillators, electric aerosol generators, sterile equipment, lighters and electronic timers, etc.

2. Method
2.1. Testing basis: Item 2.1.3.4 of "Technical Specifications for Disinfection" (2002 Edition).
2.2. Preparation of bacterial suspension: Wash Staphylococcus albus slants that have been cultured for 18 to 24 hours with nutrient broth as usual. After filtering with gauze, dilute it with nutrient broth to the required concentration, and add it to two aerosols on average in the generator.
2.3. Turn on the computer, start the aerosol chamber control software, turn on the air conditioning control button for air supply and return, and adjust the temperature and relative humidity of the two 20m³ aerosol chambers. Put the VIRSHA (potassium monopersulphate) test fluid with an effective chlorine content of 300mg/L into the electric aerosol generator at a spray volume of 10mL/m³, and place it behind the air chamber at a height of 1.5 meters. Tightly connect the aerosol generator to the aerosol chamber spray pipe, close the aerosol chamber door, activate the aerosol spray button of the aerosol chamber control software, and use the aerosol generator to simultaneously spray the two Spray the aerosol chamber to infect bacteria for 5 minutes. While spraying, the software automatically turns on the stirring fan and sets it to continue stirring for 10 minutes after the spray is completed. After the contamination and stirring are completed, let it stand for 5 minutes. Move the fully loaded plate sampler to a height of 1m in the center of the aerosol chamber, and conduct pre-disinfection sampling of the two aerosol chambers (sampling flow rate 28.3L/min). Turn on the power, perform aerosol disinfection on the right compartment (spray sterile distilled water on the left compartment), and after the predetermined time, perform post-disinfection sampling according to the previous method. Remove the plate in the sampler and place it in a 37°C incubator for 48 hours. Then count the number of bacteria and calculate the number of surviving bacteria in the air, natural death rate and killing rate under different conditions. The test was repeated three times.
2.4. Detection of ambient temperature: 21.4℃~23.0℃; relative humidity: 46%~48%.

3. Results
Under the conditions of aerosol chamber temperature of 21.4℃~23.0℃ and relative humidity of 46%~48%, put the VIRSHA (potassium monopersulphate) test fluid with an effective chlorine content of 300mg/L into the electric aerosol generator, perform aerosol spraying at a spray volume of 10mL/m³, disinfect for 60 minutes, and the killing rate of white Staphylococcus aureus in the air of the aerosol chamber is 100.00% in three repeated tests. At this time, the natural death rate of Staphylococcus albus in the air of the control aerosol chamber was below 68.54% (see attached table).

Attached table: Air disinfection simulation field test results

Experiment number	Number of control colonies before disinfection (cfu/m³)	Number of remaining colonies after disinfection (cfu/m³)	Death rate (%)
1	1802	57	96.84
2	1898	35	98.16
3	1562	28	98.21

4. Conclusion
After testing, the VIRSHA (potassium monopersulphate) test liquid with an effective chlorine content of 300mg/L was put into the electric aerosol generator, and the aerosol spray was carried out at a spray volume of 10mL/m³

for 60 minutes of disinfection. In three repeated tests, the death rate of natural bacteria in 45m³ indoor air was >90%. It complies with the disinfection qualification requirements of the "Technical Specifications for Disinfection" (2002 Edition).

Acute oral toxicity test (prototype)

Sample name: VIRSHA (potassium monopersulphate)
Test items: Acute oral toxicity test (prototype)
Sample acceptance date: February 24, 2020
Inspection completion date: June 9, 2020

1. Equipment and animals
1.1. Test sample: VIRSHA (potassium monopersulphate) (powder; active ingredients: available chlorine, available oxygen, potassium monopersulfate compound; production date/batch number: 20200208; maximum application concentration: 1:25) .
1.2. Animals: 50 KM mice, half male and half, animal age: 4 weeks, weight 18g~22g, animals provided by China Institute of Food and Drug Control (Daxing), SPF grade animals, production license number SCXK (Beijing) 2017-0005, quality certificate number 1112512011001225. The feed was provided by Beijing Keao Xieli Feed Co., Ltd., with production license number SCXK (Beijing) 2019-0003 and quality certificate number 1112622000021246. The temperature of the animal room is 19℃~26℃, and the relative humidity is 40%~70%. 12h light, 12h dark. The animals had free access to food and water.
1.3. Reagents and instruments: electronic balance (Model: ME4002E/02, No.: DL030), electronic balance (Model: ME403E/02, No.: DL031).

2. Method
2.1. Testing basis: Item 2.3.1 of "Technical Specifications for Disinfection" (2002 Edition).
2.2. Dosage grouping: Set up five dose groups of 5500mg/kg, 4271mg/kg, 3317mg/kg, 2576mg/kg, and 2000mg/kg. Each group has 10 KM mice, half male and half female.
2.3. Preparation of the test sample: Take 11.013g of the test sample, add pure water to 40mL, and mix thoroughly to obtain a high-concentration poisoning solution with a concentration of 275.0mg/mL. Dilute the solution sequentially to 213.5mg/mL, 165.8mg/mL, 128.8mg/mL, 100.0mg/mL poisoning solutions.
2.4. Poison exposure method: Animals were fasted overnight and had free access to water before exposure. The test sample was administered to the animals once by gavage, with an exposure volume of 0.2 mL/10g of body weight.
2.5. Observation: After exposure, observe and record the poisoning manifestations of the animals, the number of deaths, and the time of death. Autopsies should be performed on the dead animals and the animals that have been killed after the observation period. Observe with the naked eye. If any abnormal tissues or organs are found, further tissue analysis can be performed. Pathological examination. The observation period is 14 days.
2.6. LDso calculation method: Calculated according to Karber method.

3. Results
From the 1st day to the 4th day of exposure, 7 and 1 animals died in the 5500mg/kg and 4271mg/kg body weight groups (see attached table). Some animals showed gastric mucosal congestion during autopsy of dead animals. No abnormal clinical manifestations were found in other surviving animals during the observation period. After the observation period, the animals were sacrificed and autopsied. No abnormalities were found in the major organs by naked eye observation.

Appendix KM mouse acute oral toxicity test results

Dose(mg/kg)	Number of female animals	Number of male animals	Number of dead female animals	Number of dead male animals	Mortality rate(%)
2000	5	5	0	0	0
2567	5	5	0	0	0
3317	5	5	0	0	0
4271	5	5	0	1	10
5500	5	5	4	3	70

4. Conclusion
After testing, VIRSHA (potassium monopersulphate), the LD_{50} value of the test sample in this acute oral toxicity test to KM mice is 5098mg/kg body weight, and the 95% confidence limit is 4679mg/kg-5555mg/kg body weight. The toxicity rating is actually non-toxic.

Acute oral toxicity test (5 times application solution)

Sample name: VIRSHA (potassium monopersulphate)
Test items: Acute oral toxicity test (5 times application solution)
Sample acceptance date: February 24, 2020
Inspection completion date: June 9, 2020

1. Equipment and animals
1.1. Test sample: VIRSHA (potassium monopersulphate) (powder; active ingredients: available chlorine, available oxygen, potassium monopersulfate compound; production date/batch number: 20200208; maximum application concentration: 1:25) .
1.2. Animals: 20 KM mice, half male and half female, weighing 18g~22g, provided by China Institute of Food and Drug Control (Daxing), SPF grade animals, animal production license number SCXK (Beijing) 2017-0005, qualified quality Certificate number 1112512011001225. The feed was provided by Beijing Keao Xieli Feed Co., Ltd., with production license number SCXK (Beijing) 2019-0003 and quality certificate number 1112622000021246. The temperature of the animal room is 19℃~26℃, and the relative humidity is 40%~70%. 12h light, 12h dark. The animals had free access to food and water.
1.3. Reagents and instruments: electronic balance (Model: ME4002E/02, No.: DL030), electronic balance (Model: ME403E/02, No.: DL031).

2. Method
2.1. Testing basis: Item 2.3.1 of "Technical Specifications for Disinfection" (2002 Edition).
2.2. Dose grouping: Set up a dosage group of 5000mg/kg, 20 KM mice, half male and half female.
2.3. Preparation of test sample: Take 1.004g of sample and add 5mL of pure water and mix thoroughly to obtain 5 times the application solution. Weigh 5.012g of the 5 times application solution, add pure water to 20mL and mix thoroughly to obtain a 250mg/mL venom solution.
2.4. Poison exposure method: Animals were fasted overnight and had free access to water before exposure. The test sample was administered to the animals once by gavage, with an exposure volume of 0.2 mL/10g of body weight.
2.5. Observation: After exposure, observe and record the poisoning performance of the animals, and perform autopsies on the animals after the observation period expires. If any abnormal tissues or organs are found through naked eye observation, further histopathological examination will be conducted. The observation period is 14 days.
2.6. LD_{50} calculation method: Based on "Disinfection Technical Specifications" (2002 Edition) 2.3.1.5.2 "One Maximum Test".

3. Results
No obvious abnormal symptoms or signs were found after the animals were exposed to the poison. No other abnormal manifestations or deaths occurred after 14 days of continuous observation (see attached table). After the observation period, the animals were sacrificed for autopsy, and no abnormalities were found in the major organs by naked eye observation.

Appendix KM mouse acute oral toxicity test results

Gender	Dose group(mg/kg)	Number of animals	Number of dead animals	Mortality rate(%)
Female	5000	10	0	0
Male	5000	10	0	0

4. Conclusion
After testing, VIRSHA (potassium monopersulphate) was used in an acute oral toxicity test on KM mice using 5 times' application solution. No animals died after 14 days, and the LD_{50} was >5000mg/kg body weight. The toxicity classification was considered to be actually non-toxic. Comply with the qualification requirements of "Technical Specifications for Disinfection" (2002 Edition).

Acute inhalation toxicity test

Sample name: VIRSHA (potassium monopersulphate)
Test items: Acute inhalation toxicity test
Sample acceptance date: February 24, 2020
Inspection completion date: June 9, 2020

1. Equipment and animals
1.1. Test sample: VIRSHA (potassium monopersulphate) (powder; active ingredients: available chlorine, available oxygen, potassium monopersulfate compound; production date/batch number: 20200208; maximum application concentration: 1:25) .
1.2. Animals: 20 KM mice, half male and half female, weighing 18g~22g, purchased from China Institute of Food and Drug Control (Daxing), SPF grade animals, animal production license number SCXK (Beijing) 2017-0005, qualified quality Certificate number 1112512011001225. The feed was purchased from Beijing Keao Xieli Feed Co., Ltd., the production license number is SCXK (Beijing) 2019-0003, and the quality certificate number is 1112622000021246. The temperature of the animal room is 19℃~26℃, and the relative humidity is 40%~70%. 12h light, 12h dark. The animals had free access to food and water.
1.3. Reagents and instruments: electronic balance (Model: ME4002E/02, No.: DL030), electronic balance (Model: ME403E/02, No.: DL031), inhalation exposure cabinet (Model: 300L, No.: DL040).

2. Method
2.1. Testing basis: Item 2.3.2 of "Technical Specifications for Disinfection" (2002 Edition).
2.2. Dose grouping: According to the pre-test of the test sample, the acute inhalation toxicity of the mouse exposure cabinet is LC_{50}>10000mg/m³.This test was set to 10000mg/m³, one dose group, 20 KM mice, half male and half female.
2.3. Preparation of the test sample: Weigh 3.010g of the test sample and add pure water to make the volume to 7mL as the poisoning solution.
2.4. Poisoning method: First put 20 mice into a static poisoning cabinet (volume 0.3m³), and then take 7mL of poisoning solution, add the atomizer and inject it into the poisoning cabinet for 15 minutes, and do the poisoning for 2 hours at a time.
2.5. Observation: After exposure, observe and record the poisoning manifestations of the animals, the number of deaths, and the time of death. Autopsies should be performed on the dead animals and the animals that have been killed after the observation period. Observe with the naked eye. If any abnormal tissues or organs are found, further tissue analysis can be performed pathological examination. The observation period is 14 days.
6. Evaluation index: According to 2.3.2 of "Technical Specifications for Disinfection" (2002 Edition), inhalation exposure concentration for one 2-hour period 10000mg/m³, if there is no death within 14 days, it can be determined that the LC_{50} is greater than 10000mg/m³.

3. Results
During the exposure period, the mice had symptoms of eye closure, and no animals died. No obvious abnormal symptoms or signs were seen after the animals left the poisoning cabinet. No other abnormal symptoms or animal death occurred after 14 days of continuous observation (see attached table).

Appendix KM mouse acute inhalation toxicity test results

Gender	Dose group(mg/kg)	Number of animals	Number of dead animals	Mortality rate(%)
Female	10000	10	0	0
Male	10000	10	0	0

4. Conclusion
After testing, VIRSHA (potassium monopersulphate) was subjected to an acute inhalation toxicity test on KM mice. No animals died in 14 days, LC_{50}>10000mg/m³, and the toxicity classification is actually non-toxic. Comply with the qualification requirements of "Technical Specifications for Disinfection" (2002 Edition).

A complete skin irritation test

Sample name: VIRSHA (potassium monopersulphate)
Test items: A complete skin irritation test
Sample acceptance date: February 24, 2020
Inspection completion date: May 29, 2020

1. Equipment and animals
1.1. Test sample: VIRSHA (potassium monopersulphate) (powder; active ingredients: available chlorine, available oxygen, potassium monopersulfate compound; production date/batch number: 20200208; maximum application concentration: 1:25) .
1.2. Animals: 3 Japanese big-eared white rabbits, 3 months old, female, weighing 2.0kg~2.5kg. The animals are provided by Beijing Changyang Xishan Breeding Farm. They are ordinary-level animals. Production license number is SCXK (Beijing) 2016- 0007, quality certificate number 1103292011000252. The feed was provided by Beijing Keao Xieli Feed Co., Ltd., with production license number SCXK (Beijing) 2019-0003 and quality certificate number 1112622000021106. The temperature of the animal room is 19℃~26℃, and the relative humidity is 40%~70%. 12h light, 12h dark. The animals had free access to food and water.

2. Method
2.1. Testing basis: Item 2.3.3.3.1 of "Technical Specifications for Disinfection" (2002 Edition).
2.2. Preparation of the test sample: Weigh 1.004g of the sample, add 5mL of pure water to fully dissolve and mix, and prepare a 5-fold application solution.
2.3. Exposure method: 24 hours before the test, remove hair from both sides of the spine on the back of the Japanese big-eared white rabbit, covering an area of about 3cm×3cm. The next day, 0.5mL of the prepared 5x application solution was directly dropped on the left side of the completely depilated skin with an area of 2.5cm× 2.5cm, then covered with a layer of non-irritating plastic film, and then fixed with non-irritating tape. The right side is used as a blank control. 4 hours after application, wash with warm water to remove residual test samples.
2.4. Observation and evaluation: Observe local skin reactions and score 1h, 24h and 48h after removal of residual test samples. The scoring standards and irritation intensity classification refer to Section 2.3.3 "Skin Irritation" of "Technical Specifications for Disinfection" (2002 Edition) Table 2-11 and Table 2-12 in "Test".

3. Results
After removing the residual test sample, animal No. 1003 developed barely visible erythema at 1 hour and obvious erythema at 24 and 48 hours (see attached table for scores).

Attached table: Results of a complete skin irritation reaction score of Japanese big-eared white rabbits

Animal Number			1001	1002	1003	Integral Mean
Gender			f	f	f	
1h	Sample	Erythema	0	0	1	0.33
		Edema	0	0	0	
		Total Score	0	0	0	
	Control	Erythema	0	0	0	0
		Edema	0	0	0	
		Total Score	0	0	0	
24h	Sample	Erythema	0	0	2	0.67
		Edema	0	0	0	
		Total Score	0	0	0	
	Control	Erythema	0	0	0	0
		Edema	0	0	0	
		Total Score	0	0	0	

48h	Sample	Erythema	0	0	2	0.67
		Edema	0	0	0	
		Total Score	0	0	0	
	Control	Erythema	0	0	0	0
		Edema	0	0	0	
		Total Score	0	0	0	

4. Conclusion

After testing, VIRSHA (potassium monopersulphate) was used for a complete skin irritation test on Japanese big-eared white rabbits using 5 times the application solution. The highest skin irritation index was 0.67 (0.5~<2.0), and the irritation intensity was mildly irritating. Comply with the qualification requirements of "Technical Specifications for Disinfection" (2002 Edition).

Acute Eye Irritation Test

Sample name: VIRSHA (potassium monopersulphate)
Test items: acute eye irritation test
Sample acceptance date: February 24, 2020
Inspection completion date: June 05, 2020

1. Equipment and animals
1.1. Test sample: VIRSHA (potassium monopersulphate) (powder; active ingredients: available chlorine, available oxygen, potassium monopersulfate compound salt; production date/batch number: 20200208; maximum application concentration: 1:25) .
1.2. Animals: 3 Japanese big-eared white rabbits, female, weighing 2.0kg~2.5kg, provided by Changyang Xishan Breeding Farm in Beijing, ordinary grade animals, animal production license number SCXK (Beijing) 2016-0007, qualified quality Certificate number 1103292011000252. The feed was provided by Beijing Keao Xieli Feed Co., Ltd., with production license number SCXK (Beijing) 2019-0003 and quality certificate number 1112622000021106. The temperature of the animal room is 19℃~26℃, and the relative humidity is 40%~70%. 12h light, 12h dark. The animals had free access to food and water.

2. Method
2.1. Testing basis: Item 2.3.4 of "Technical Specifications for Disinfection" (2002 Edition).
2.2. Preparation of the test sample: Weigh 1.007g of the sample, add 5mL of pure water to fully dissolve and mix, and prepare a 5-fold application solution.
2.3. Exposure method: absorb 0.1 mL of 5 times the application solution of the test sample, and drop it into the conjunctival sac of the left eye of the Japanese big-eared white rabbit. After instilling the test sample, passively close the eye for 4 seconds, and then use physiological medicine for 30 seconds. Rinse with salt water. Normal saline was instilled in the right eye as a normal control.
2.4. Observation and evaluation: At 1h, 24h, 48h, 72h, and 7d after eye drops, the damage and recovery of the conjunctiva, iris and cornea of Japanese big-eared white rabbits were visually observed. If no irritation reaction occurs within 72 hours, or the eye irritation reaction completely recovers on the 7th or 14th day, the test will be terminated early. The scoring standards and irritation intensity classification refer to Table 2-13 and Table 2-14 in Section 2.3.4 "Acute Eye Irritation Test" of the "Technical Specifications for Disinfection" (2002 Edition).

3. Results
After exposure, no abnormalities were found in the eyes of the animals' exposed and control sides at 1h, 24h, 48h, and 72h (see attached table), so the test was terminated early.

Attached table: Japanese big-eared white rabbit acute eye irritation (within 72 hours) reaction score results

Serial Number		1h		24h		48h		72h		Average Rating	
	Damage Performance	Sample	Control	Sample	Control	Sample	Control	Sample	Control	Sample	Control
1001	Corneal Damage	0	0	0	0	0	0	0	0	0	0
	Iris Damage	0	0	0	0	0	0	0	0	0	0
	Conjunctival Hyperemia	0	0	0	0	0	0	0	0	0	0
1002	Conjunctival Edema	0	0	0	0	0	0	0	0	0	0
	Corneal Damage	0	0	0	0	0	0	0	0	0	0
	Iris Damage	0	0	0	0	0	0	0	0	0	0
	Conjunctival Hyperemia	0	0	0	0	0	0	0	0	0	0
1003	Conjunctival Edema	0	0	0	0	0	0	0	0	0	0
	Corneal Damage	0	0	0	0	0	0	0	0	0	0
	Iris Damage	0	0	0	0	0	0	0	0	0	0

	Conjunctival Hyperemia	0	0	0	0	0	0	0	0	0	0

*Average rating is the sum of 24h, 48h and 72h divided by the number of observation periods 3.

4. Conclusion

After testing, VIRSHA (potassium monopersulphate), using 5 times the application solution for an acute eye irritation test on Japanese white rabbits, found that the corneal damage was <1, the iris damage was <1, the conjunctival congestion was <2, and the conjunctival edema was <2. Injury type It is non-irritating. Comply with the qualification requirements of "Technical Specifications for Disinfection" (2002 Edition).

Mouse bone marrow polychromatic erythrocyte micronucleus test

Sample name: VIRSHA (potassium monopersulphate)
Test items: Mouse bone marrow polychromatic erythrocyte micronucleus test
Sample acceptance date: February 24, 2020
Inspection completion date June 22, 2020

1. Equipment and animals
1.1. Test sample: VIRSHA (potassium monopersulphate) (powder; active ingredients: available chlorine, available oxygen, potassium monopersulfate compound salt; production date/batch number: 20200208; maximum application concentration: 1:25) .
1.2. Animals: 50 KM mice, half male and half female, weighing 25g~30g, provided by China Institute for Food and Drug Control (Daxing), SPF grade animals, animal production license number SCXK (Beijing) 2017-0005, quality qualified Certificate number 1112512011001564. The feed was provided by Beijing Keao Xieli Feed Co., Ltd., with production license number SCXK (Beijing) 2019-0003 and quality certificate number 1112622000021246. The temperature of the animal room is 19℃~26℃, and the relative humidity is 40%~70%. 12h light, 12h dark. The animals had free access to food and water.
1.3. Reagents and instruments: cyclophosphamide (manufacturer: Macklin, batch number: C10478481), electronic balance (model: ME4002E/02, number: DL030), electronic balance (model: ME403E/02, number: DL031) biological microscope (model : OlympusCX41, No.: DL001).

2. Method
2.1. Test basis: Item 2.3.8.4 of "Technical Specifications for Disinfection" (2002 Edition).
2.2. Animal grouping: The acute oral toxicity test of this sample in mice resulted in an LD_{50} of 5098 mg/kg. The test was divided into three dose groups, namely 2549 mg/kg, 1020 mg/kg, and 255 mg/kg, and a cyclophosphamide positive control group was set up. (40mg/kg), pure water negative control group, 10 mice in each group, half male and half female.
2.3. Sample preparation: Weigh 2.549g of the sample and add pure water to adjust the volume to 20mL to prepare a 127.5mg/mL poisoning solution. The solution is diluted sequentially into 51.0mg/mL and 12.8mg/mL poisoning solutions. Weigh 0.020g of cyclophosphamide, add physiological saline to 10mL, mix well and set aside.
2.4. Exposure method: Animals were exposed to the poison by oral gavage for 30 hours, that is, the interval between two exposures was 24 hours, and the exposure volume was 0.2 mL/10 g of body weight. Samples were collected 6 hours after the second exposure.
2.5. Obtain materials and make films: kill the animals by cervical dislocation. Take 2 femurs, peel off the muscles, and wipe away the blood. Cut off both ends of the femur to expose the medullary cavity. Aspirate 0.1 mL of calf serum and flush the bone marrow cavity. Use regular smear with rinse solution and let dry naturally.
2.6. Staining: Fix the dried smear in methanol for 10 minutes, stain with Giemsa stain for 15 minutes, then rinse with PBS solution (pH 6.8) and dry.
2.7. Observation: Select an area where cells are evenly distributed, complete, and properly colored, and count the number of polychromatic erythrocytes (PCE) containing micronuclei under an oil microscope. Count 1,000 PCEs per animal. The micronucleus cell rate refers to the number of PCEs containing micronuclei in 1,000 PCEs, expressed in parts per thousand. Count 200 PCE and count the mature red blood cells (NCE) seen at the same time to find the ratio of PCE/NCE.
2.8. Statistical processing: Data were statistically analyzed using x^2 test and variance analysis, and completed with SPSS software.

3. Results
Test samples induced micronucleus in mouse bone marrow polychromatic erythrocytes. Compared with the negative control group, there was no significant difference in the micronucleus rate of animal bone marrow polychromatic erythrocytes in each dose group ($p>0.05$); cyclophosphamide positive control Compared with the negative control group, there was a very significant difference ($p<0.01$). The PCE/NCE ratio of animal bone marrow in each dose group was greater than 0.1, indicating that the test sample had no obvious inhibitory effect on bone marrow, and there was no significant difference in the PCE/NCE ratio of the sample in each dose group compared with the negative control group ($p>0.05$) (See Schedule). During the administration period, no obvious abnormality was found in the animals in each dose group.

Attached table: Mouse bone marrow polychromatic erythrocyte micronucleus test results

Group	Dose (Mg/Kg)	Number Of Animals (Only)	Number Of Pces Inspected (Number)	Number Of Pces Containing Microcores	Micronucleus Cell Rate (%, x±s)	Pce/Nce (x±s)

				(Number)		
Test Sample Group	225	10	10000	4	0.40±0.52	1.29±0.13
	1020	10	10000	2	0.20±0.42	1.28±0.11
	2549	10	10000	3	0.30±0.67	1.28±0.13
Negative Control Group	0	10	10000	2	0.20±0.42	1.25±0.10
Positive Control Group	40	10	10000	130	13.00±3.06**	1.03±0.11**

Note: Micronucleus cell rate (%) and PCE/NCE are calculated in mice and expressed as mean ± standard deviation.
**p<0.01, compared with negative control group.

4. Conclusion
After testing, VIRSHA (potassium monopersulphate) has no micronucleogenic effect on mouse bone marrow polychromatic erythrocytes. Comply with the qualification requirements of "Technical Specifications for Disinfection" (2002 Edition).

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Seal for inspection and testing

Final Review Date: Jan 4, 2021