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Test method for efficacy of antimicrobials - Aerobic bacteria

杀生剂能效的评价方法 异养菌

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Foreword

Appendix A and Appendix B of this Standard are normative.

This Standard was proposed by China Petroleum and Chemical Industry Federation.

This Standard shall be administrated by Water Treatment Sub-committee of National Standardization Technical Committee (SAC/TC 63/SC 5).

Responsible drafting organizations of this Standard: Shanghai Future Enterprises Limited, and CNOOC Tianjin Chemical Research and Design Institute.

Main drafters of this Standard: Liu Xin, Zhang Quan, Shao Hongqian, and Zhu Chuanjun.

Test method for efficacy of antimicrobials – Aerobic bacteria

1 Scope

This Standard specifies the test method for the sterilization efficacy of aerobic bacteria by using antimicrobial in cooling water system.

This Standard applies to the test for the sterilization efficacy of oxidizing and non-oxidizing antimicrobial to the aerobic bacteria.

2 Normative references

The provisions in following documents become the provisions of this Standard through reference in this Standard. For dated references, the subsequent amendments (excluding corrections) or revisions do not apply to this Standard, however, parties who reach an agreement based on this Standard are encouraged to study if the latest versions of these documents are applicable. For undated references, the latest edition of the referenced document applies.

GB/T 603 Chemical reagent - Preparations of reagent solutions for use in test methods (GB/T 603-2002, ISO 6353-11982, NEQ)

GB/T 6682 Water for analytical laboratory use - Specification and test methods (GB/T 6682-2008, ISO 3696: 1987, MOD)

GB/T 14643.1 Industrial circulating cooling water - Examination of bacteria formed deposits - Standard of plate count

3 Summary of methods

In the sample that contains a certain amount of aerobic bacteria. ADD antimicrobials quantitatively. SIMULATE antimicrobial's use conditions (water quality, pH value, etc.). DETERMINE the amount of aerobic bacteria survived in the prescribed time period and COMPARE it with the amount of aerobic bacteria at the beginning of test. CALCULATE the killing rate of this

cooling water, the selection range of concentration of the antimicrobial to be added shall be in accordance with the characteristics and use conditions of the antimicrobial. CHOOSE several concentration grades in a certain range to conduct test.

7.1.3 To make dosing operation more convenient, the original fluid of antimicrobial to be tested shall be diluted with water before the test. The degree of dilution shall be based on - after the diluted solution of antimicrobial is added to the sample, it just reaches the concentration needed in test (the ratio of the volume of diluted antimicrobial and the volume of sample to be added shall not exceed 1:100). The diluted solution of antimicrobial shall be used immediately after it is diluted.

7.2 Test procedure

7.1.1 General test

7.1.1.1 This test is used to choose the appropriate type and appropriate dosing concentration of antimicrobial.

7.2.1.2 TAKE 200 mL of sample prepared in 6.7 of which the content of aerobic bacteria is 10^5 cfu/mL~10^7 cfu/mL. ADD it to the erlenmeyer flasks (the specific amount of flask is determined by the total amount and concentration of test antimicrobial in same batch). STUFF with cotton.

7.1.1.3 DETERMINE the amount of aerobic bacteria survived in the water samples according to GB/T 14643.1. That is the initial amount of bacteria of the test of same batch.

7.1.1.4 NUMBER the erlenmeyer flasks. ADD the antimicrobial dilution in 7.1.3 to each flask correspondingly. SHAKE them well.

7.2.1.5 PLACE the entire erlenmeyer flask in a thermostatic biochemical incubator at (29 ± 1) ºC to culture.

7.2.1.6 DETERMINE the total amount of aerobic bacteria in the erlenmeyer flask at the 4th hour, 8th hour, 12th hour, 16th hour, 20th hour, 24th hour (the specific test time is determined according to the characteristics and use

of antimicrobial to conduct test.

B.2.2 Preparation of antimicrobial solutions with different concentrations

DELUTE the original fluid of antimicrobial with the water for test. The degree of dilution shall be based on - to make the diluted liquor of antimicrobial just reaching the concentration needed by the test, after adding quantitative culture medium.

B.2.3 NUMBER the petri dishes. INDICATE the name and concentration of the selected antimicrobials.

B.2.4 USE sterile pipette to transfer 1 mL of diluted liquor of antimicrobial in B.2.2. ADD it to the culture dish with corresponding number. And quantitatively INJECT 10mL~15mL of sterile culture medium that is cooled to about (45 ± 1) ºC into culture dish immediately. SHAKE them well. COOL it for spare use.

B.2.5 USE micro-injector to take 10 μL of sample of 6.7. PLACE it at the center of bacteria-bearing culture medium plate (prepared in B.2.4). COVER it with lid.

B.2.6 PLACE B.2.5 at the appropriate temperature for a certain period of time (determined according to different test conditions). According to the growth condition of the sample on the plate containing antimicrobial, DETERMINE the minimum inhibitory concentration of antimicrobial to the sample, so as to judge the size of sterilization efficacy of antimicrobial.

Note: The minimum inhibitory concentration method is a quantitative method, it can better reflect the size of the sterilization efficacy of antimicrobial, and it is convenient to compare the sterilization efficacy of different antimicrobials. However, the determination data from different sources are often different, some differences are great, this kind of difference is so difficult to avoid that even in the tests of different batches in same laboratory.

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