NATIONAL STANDARD OF THE

PEOPLE'S REPUBLIC OF CHINA

GB 4789.2-2022

National food safety standard - Microbiological examination of food: Aerobic plate count

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Foreword

This Standard replaces GB 4789.2-2016 "National food safety standard - Food microbiological examination - Aerobic plate count".

Compared with GB 4789.2-2016, the main changes of this Standard are as follows.

- Add Appendix B;
- Modify the equipment and materials;
- Modify the medium and reagents;
- Modify the examination procedure;
- Modify the operation steps;
- Modify Appendix A.

National food safety standard - Microbiological examination of food. Aerobic plate count

1 Scope

This Standard specifies the method for the determination of aerobic plate count in food.

This Standard applies to the determination of aerobic plate count in food.

2.1 Aerobic plate count

The total number of microbiological colonies formed in per g (mL) of test sample, which is obtained after the food sample under test is processed and cultured under certain conditions (such as culture medium, culture temperature, and incubation time, etc.).

3 Equipment and materials

In addition to the routine sterilization and culture equipment in the microbiology laboratory, other equipment and materials are as follows.

a) Constant-temperature incubator.

b) Refrigerator.

c) Thermostat.

d) Balance. Sensitivity is 0.1 g.

e) Homogenizer.

f) Oscillator.

g) Sterile straw. 1 mL (with 0.01 mL scale), 10 mL (with 0.1 mL scale) or micropipette and tip.

h) Sterile conical flask. Capacity is 250 mL and 500 mL.

i) Sterile petri dish. Diameter is 90 mm.

j) pH meter or pH colorimetric tube or precision pH test paper.

k) Magnifying glass or/and colony counter.

4.2 Test piece of aerobic plate count. It shall meet the quality control requirements of

plate count agar medium in GB 4789.28.The main nutrients are consistent with the formula of plate count agar medium.

5 Examination procedure

The examination procedure for aerobic plate count is shown in Figure 1.

6.1.1 Solid and semi-solid sample. Weigh 25 g of sample; place it in a sterile

homogeneous cup filled with 225 mL of sterile phosphate buffer or sterile normal saline. Homogenize at 8000 r/min~10000 r/min for 1 min~2 min. Or put it into a sterile homogeneous bag containing 225 mL of diluent. Use a slap-type homogenizer to beat for 1 min~2 min, to make a 1.10 sample homogenate.

Test sample25 g (mL) of sample + 225 mL of diluent,homogenized10-fold serial dilution Culture Add 15 mL~20 mL of plate count agar

medium to each dish and mix well Select 1~3 sample homogenates of

appropriate dilution; add 1 mL of each to asterile petri dish Count the number of colonies

on each plate Calculate the aerobic plate count Report Culture at 36 °C+/-1 °C for 48 h+/-2 h. Aquatic

products are cultured at 30 °C+/-1 °C for 72 h+/-3 h

6.1.2 Liquid sample. Use a sterile straw to pipette 25 mL of sample; place it in a sterile

conical flask containing 225 mL of sterile phosphate buffer or sterile normal saline (an appropriate number of sterile glass beads can be preset in the bottle); and mix thoroughly. Or put it into a sterile homogeneous bag containing 225 mL of diluent. Use a slap-type homogenizer to beat for 1 min~2 min, to make a 1.10 sample homogenate. When the result is required to be the aerobic plate count per g of sample, operate according to 6.1.1.

6.1.3 Use a 1 mL sterile straw or micropipette to draw 1 mL of the 1.10 sample

homogenate; along the tube wall, slowly inject it into a sterile test tube containing 9 mL of diluent (be careful not to touch the surface of the diluent with the pipette or the tip). Oscillate and mix on an oscillator, to make a 1.100 sample homogenate.

6.1.4 According to the operation in 6.1.3, prepare a 10-fold serial dilution of the sample

homogenate. For each incremental dilution, use a new 1 mL sterile straw or tip.

6.1.5 According to the estimation of the contamination status of sample, select 1 to 3

sample homogenates with appropriate dilution (liquid samples can include stock solution). Pipette 1 mL of the sample homogenate into a sterile petri dish; make two petri dishes for each degree of dilution. At the same time, respectively pipette 1 mL of blank diluent; add it to two sterile petri dishes as blank control.

46 °C~50 °C (which can be kept in a thermostat at 48 °C+/-2 °C) into a petri dish; rotate

the petri dish to make it evenly mixed.

36 °C+/-1 °C for 48 h+/-2 h. Aquatic products are cultured at 30 °C+/-1 °C for 72 h+/-3 h. If

the sample may contain colonies that spread and grow on the surface of the agar medium, it is possible to cover the surface of the solidified agar medium with a thin layer of plate count agar medium (about 4 mL). After solidification, turn the plate over and culture.

6.2.2 If the test piece of aerobic plate count is used, it shall be operated in accordance

with the relevant technical regulations provided for the test piece.

6.3.3 When one of the plates has larger flaky colonies, it is not suitable to use it. The

plate without larger flaky colonies shall be used as the colony count of the degree of dilution. If the flaky colonies are less than half of the plate, and the colonies in the remaining half are evenly distributed, it is possible to calculate the number of the half plate and multiply it by 2, to represent the number of colonies on one plate.

6.3.4 When there is a chain growth with no clear boundary between the colonies on the

plate, count each single chain as a colony.