SimPlate® *Total Plate Count*

Introduction

SimPlate® for Total Plate Count (TPC) method is used for the detection and quantification of the total aerobic microorganisms in food. The medium/sample mixture is dispensed into a SimPlate® device and incubated for 24-28 h. The total aerobic plate count is determined by counting the fluorescent wells and referring to the SimPlate® Conversion Table. The SimPlate® device is packaged separately.

Single Test Medium

Kit Components

100 individually-packaged dehydrated TPC medium containers.

Multiple Test Medium

Kit Components

50 multi-test dehydrated TPC medium containers. Each container is sufficient for 10 tests.

A. Sample Preparation

a. Weigh 50 g of sample into 450 mL of sterile diluent [Butterfield's phosphate buffer (AOAC Method) or peptone salt solution (ISO Method)]. This is a 10-fold dilution. Masticate or blend to homogenize.

b. If an alternate sample size is specified in your testing procedure or standard, prepare a 10% weight to volume suspension.

c. If necessary, prepare 10-fold serial dilutions appropriate for the anticipated population of the sample.

B. Test Procedure (FOR SINGLE TEST)

For 1.0 mL sample size

a. Resuspend powdered medium with 9.0 mL of sterile deionized water containing 1 mL of Supplement A per 100 mL. Add 1.0 mL of sample and mix well. **DO NOT** count this reconstitution as a dilution.

For 0.1 mL sample size

b. Resuspend powdered medium with 9.9 mL of sterile deionized water containing 1 mL of Supplement A per 100 mL. Add 0.1 mL of sample and mix well. This is an additional 10-fold dilution.

Note: The final volume of sample/medium mixture in the container should be 10 ±0.2 mL. Mix well.

c. Remove the lid from the SimPlate® device and pour the sample/ medium mixture onto the center of the plate (Figure 1). Immediately replace the lid.

B. Test Procedure (FOR MULTIPLE TESTS)

a. Empty contents of one container into 100 mL of sterile deionized water containing 1 mL of Supplement A per 100 mL. Shake to completely dissolve.

b. Remove the lid from the SimPlate® device. If prepared sample size is 1.0 mL, pipette it onto the center of the device (Figure 2). Overlay the sample with 9.0 mL of medium. **DO NOT** count this media addition as a dilution.

c. For 0.1 mL of prepared sample, overlay it with 9.9 mL of medium: this is an additional 10-folddilution.
Note: The final volume of sample/medium mixture on the plate should be 10 ±0.2 mL. Immediately replace the lid.

d. Gently swirl to distribute the sample/medium mixture into all the wells (Figure 3). The plate may be held with both hands and tilted slightly to help distribute the liquid into the wells.

e. Pour off excess medium by holding the lid against the plate on either side of the sponge cavity. Tip the plate toward you to allow liquid to drain into the sponge (Figure 4).Do not be concerned if partially filled

wells are present. Wells containing partial volume of liquid will turn positive in the presence of viable bacteria.

f. **DO NOT** invert the SimPlate® device. If testing in accordance with AOAC®/BAM/USDA methods, incubate in the dark for 24 to 28 h at 35 º ±1 ºC (32 º ±1 ºC for dairy products). If testing in accordance with EN/ISO standards, incubate in the dark for 24 to 28 h at 30 º ±1 ºC (for all products).

Figure 1

For single test, pour sample/ medium mixture onto the center of the plate.

Figure 2

For multiple tests, pipette sample onto center of plate. Add rehydrated medium to make a final volume of 10 ±0.2 mL.

Figure 3

Cover plate, gently swirl to distribute the sample into all of the wells.

Figure 4

Tap plate GENTLY on a hard surface to remove air bubbles.

Figure 5

Holding the cover, tip the plate toward you to allow liquid to drain.

C. Reading and Interpretation of Results

a. After incubation, count the number of wells showing blue fluorescence by holding a UV light (366 nm) approximately 15–30 cm (6–12 in) above the SimPlate® device.

b. To determine the population per plate, perform the following calculations:

1. Count the number of positive (blue fluorescence) wells in the plate.
2. Use the SimPlate® Conversion Table to determine the total number of microorganisms per plate

c. To calculate the number of **microorganisms per g (mL),** multiply the count in **C(b)2** by the appropriate dilution factor (sections **A** and **B**)

CI. Product and Storage Information

a. Store dehydrated medium away from direct light between 2–30 °C.

b. DO NOT use expired medium.

c. Store containers of reconstituted medium between 15 and 25 °C and use within 12 h.

d. Handle and dispose of incubated medium in a decontamination container and sterilize according to Good Laboratory Practices.

Manufacturing Entity

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