



Official Methods of Analysis of AOAC INTERNATIONAL (22nd Edition)

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CHAPTER

17.2.07 AOAC Official Method 990.12 Aerobic Plate Count in Foods: Dry Rehydratable Film Method Neogen® Petrifilm® Aerobic Count Plate

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First Action 1990

Final Action 1994

Results of the interlaboratory study supporting acceptance of the method:

Flour: $s_T = 0.225$; $s_R = 0.246$; $RSD_T = 5.3\%$; $RSD_R = 5.8\%$

Nuts: $s_T = 0.272$; $s_R = 0.674$; $RSD_T = 7.4\%$; $RSD_R = 18.4\%$

Shrimp: $s_T = 0.540$; $s_R = 0.615$; $RSD_T = 9.8\%$; $RSD_R = 11.1\%$

Spice: $s_T = 0.274$; $s_R = 0.303$; $RSD_T = 6.0\%$; $RSD_R = 6.6\%$

Turkey: $s_T = 0.278$; $s_R = 0.348$; $RSD_T = 5.3\%$; $RSD_R = 6.6\%$

Vegetables: $s_T = 0.310$; $s_R = 0.454$; $RSD_T = 6.3\%$; $RSD_R = 9.2\%$

A Principle

See 989.10A (see 17.3.03).

B Apparatus

See 989.10B (a) and (c)–(e) (see 17.3.03).

C Reagent

Butterfield's Phosphate Buffered Dilution water.—To prepare stock solution, dissolve 34 g KH_2PO_4 in 500 mL H_2O , adjust to pH 7.2 with 1 M NaOH (ca 175 mL), and dilute to 1 L with water. To prepare buffered water for dilutions, dilute 1.25 mL stock solution to 1 L with boiled and cooled water. Autoclave 15 min at 121°C.

D Preparation of Test Suspension

See 966.23B (see 17.2.01).

E Determination

Place dry-film aerobic count plate on flat surface. Lift top film and inoculate 1 mL test suspension onto center of film base. Carefully place top film down on inoculum. Distribute suspension over prescribed growth area with downward pressure in center of plastic spreader device (recessed side down). Leave plate undisturbed 1 min to permit gel to solidify. Incubate plates 4.8 ± 3 h at $35 \pm 1^\circ\text{C}$.

In incubator, place plates in horizontal position, clear side up, in stacks not exceeding 20 units. Count plates promptly after incubation period. After incubation is complete, plates may be stored frozen ($\leq -15^\circ\text{C}$) up to 7 days. Avoid this as a routine practice.

Use standard colony counter for counting purposes. Magnifier-illuminator may also be used to facilitate counting. Colonies stain in various shades of red. Count all colonies in countable range (30–300 colonies).

To compute bacterial count, multiply total number of colonies per plate (or average number of colonies per plate if counting duplicate plates of same dilution) by reciprocal of dilution used. When counting colonies on duplicate plates of consecutive dilutions, compute mean number of colonies for each dilution before determining average bacterial count. Estimated counts can be made on plates with >300 colonies and should be reported as estimated counts. In making such counts, circular growth area can be considered to contain ca twenty 1 cm squares. To isolate colonies for further identification, lift top film and pick colony from gel.

Reference:

JAOAC 73, 242(1990)

Revised: March 2002; November 2023 (brand update and minor editorial corrections)