

STANDARD METHODS AGAR (7157)

Intended Use

Standard Methods Agar is used for the enumeration of bacteria in water, wastewater, food, and dairy products. This formula conforms to American Public Health Association¹ (APHA), and Association of Official Analytical Chemists² (AOAC).

Product Summary and Explanation

Standard Methods Agar was developed by Buchbinder, Baris, and Goldstein³ in 1953 at the request of the American Public Health Association. Results showed that a dehydrated milk-free medium containing 0.25% Yeast Extract, 0.5% Tryptone, 0.1% Dextrose, and 1.5% Agar per liter approximated the productivity of Tryptone Glucose Extract Agar with added milk. Buchbinder et al.⁴ recommended that a dehydrated culture medium be used in preparing the standard plate count medium rather than preparing the medium from ingredients. Standard Methods Agar as originally suggested by Buchbinder et al.⁴

Standard Methods Agar is also referred to as Plate Count Agar and Tryptone Glucose Yeast Agar. This formula is specified in standard method procedures.^{1,2,5-7}

Principles of the Procedure

Enzymatic Digest of Casein and Yeast Extract provide the carbon and nitrogen sources required for growth of a wide variety of organisms. Dextrose is a source of fermentable carbohydrate (energy source). Agar is the solidifying agent.

Formula / Liter

Enzymatic Digest of Casein 5 g	g
Yeast Extract	ġ
Dextrose (Glucose)1	g
Agar*15 g	g
* 9 – 18 g according to gel strength	-

Final pH: 7.0 ± 0.2 at 25°C

Formula may be adjusted and/or supplemented as required to meet performance specifications.

Precaution

1. For Laboratory Use.

Directions

- 1. Suspend 23.5 g of the medium in one liter of purified water.
- 2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
- 3. Autoclave at 121°C for 15 minutes.

Quality Control Specifications

Dehydrated Appearance: Powder is homogeneous, free flowing, and light beige.

Prepared Appearance: Prepared medium is trace to slightly hazy, and light beige to medium amber.

Expected Cultural Response: Raw milk dilutions were prepared and tested following the standardized test method as outlined in Standard Methods for the Microbiological Examination of Dairy Products, incubated at $32 \pm 1^{\circ}$ C, and examined for growth at 48 hours.

Test Sample	Expected Results
Unpasteurized (raw) milk	t-value < 2.70



Test Procedure

- 1. Perform serial dilutions on samples (food, water) to be tested using the heterotrophic (standard) plate count method. Select dilutions that will yield plates with counts of 30 300 colonies.
- 2. Dispense a portion of each test dilution (e.g., 0.1 mL, 1.0 mL) into separate test dilutions.
- 3. Add 10 12 mL of tempered (45°C) Standard Methods Agar to petri dishes containing test dilutions.
- 4. Swirl the dishes to thoroughly mix the agar and test dilution.
- 5. Allow plates to cool and solidify.
- 6. Incubate at $32 \pm 1^{\circ}$ C for 48 hours.

<u>Results</u>

Count colonies on all plates containing 30 - 300 colonies. Calculate bacterial count per milliliter of sample by multiplying the average number of colonies per plate by the reciprocal of the dilution used. Report the count as CFU/mL.

Storage

Store sealed bottle containing the dehydrated medium at 2 - 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping container tightly closed.

Expiration

Refer to expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if the appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitation of the Procedure

Due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.

Packaging

Standard Methods Agar	Code No.	7157A	500 g
-		7157B	2 kg
		7157C	10 kg

References

- 1. **Marshall, R. T. (ed.).** 1993. Standard methods for the microbiological examination of dairy products, 16th ed. American Public Health Association, Washington, D.C.
- 2. Cunnif, P. (ed.). 1995. Official methods of analysis AOAC International, 16th ed. AOAC International, Arlington, VA.
- 3. Buchbinder, L., Y. Baris, and L. Goldstein. 1953. Further studies on new milk-free media for the standard plate count of dairy products. Am J. Public Health 43:869-872.
- 4. Buchbinder, L., Y. Baris, E. Alff, E. Reynolds, E. Dillon, V. Pessin, L. Pincus, and A. Strauss. 1951. Studies to formulate new media for the standard plate count of dairy products. Pub. Health Rep. 66:327-340.
- 5. Vanderzant, C., and D. F. Splittstoesser (eds.). 1992. Compendium of methods for the microbiological examination of foods, 3rd ed. American Public Health Association, Washington, D.C.
- 6. **Greenberg, A. E., L. S. Clesceri, and A. D. Eaton (eds.).** 1992. Standard methods for the examination of water and wastewater, 18th ed. American Public Health Association, Washington, D.C.
- 7. www.fda.gov/Food/ScienceResearch/LaboratoryMethods/BacteriologicalAnalyticalmanualBAM/default.htm.

Technical Information

Contact Acumedia Manufacturers, Inc. for Technical Service or questions involving dehydrated culture media preparation or performance at (517)372-9200 or fax us at (517)372-2006.

