STANDARD METHODS AGAR

GAMMA IRRADIATED

PRODUCT:

Plate Media:

Standard Methods Agar	P8680IC (Single Wrap), P8140 (Triple Wrap, Contact plate,16 mL, 60 mm)

PURPOSE:

Standard Methods Agar is a medium used for the enumeration of bacteria from water, wastewater, dairy products, and foods.

PRINCIPLE:

Standard Methods Agar is a modification of the medium designed by Bowers and Hucker[1] in 1944. It is now prepared by the specifications of the American Public Health Association and is recommended for use in the performance of standard plate counts on dairy products.[3]

Peptone, yeast extract, and glucose provide the nutritional requirements needed for growth by most bacteria. The medium has the advantage of providing more clarity than the original medium that contained skim milk. This medium is also differential in that it allows detection of organisms that are capable of breaking down casein by proteolytic action. Colonies have an increased size and permit an easier means of enumeration and interpretation.

Gamma irradiation of the packaged plates allows this product to be used in critical environments where introduction of contaminants is not desired. With **triple wrap**, aseptic removal of the inner packaging from the outer wrap allows the product to be moved into a clean room environment without the contaminated outer wrap. Product in the third inner wrap eliminates the need to aseptically rewrap the product for transport or storage and possibly contaminating the medium. **Single Wrap** provides the user an alternative in irradiated plates for those with less extensive isolation requirements. It utilizes the properties of **Tyvek®** (vaporpermeable, water-, chemical-, puncture-, tear- and abrasion-resistant, will not tear under heavier than normal useage) as the outer wrap.

FORMULA:

Approximate, per liter deionized filtered water.

Pancreatic Digest of Casein	5.0 g
Yeast Extract	2.5
Dextrose	1.0
Agar	15.0

Final pH 7.0 ± 0.2 at 25°C

PRECAUTIONS:*

For in vitro diagnostic use. Observe approved biohazard precautions.

Storage: Upon receipt store at 2-8°C away from direct light. Media should not be used if there are signs of contamination, deterioration (shrinking, cracking, or discoloration), or if the expiration date has passed. Do not open outer wrapping until ready to use. Media can be inoculated up to the expiration date and incubated for the appropriate incubation period.

Limitations: This medium becomes cloudy at incubation temperatures higher than 35-45°C due to decreased solubility of calcium salts.

When the integrity of the seal or outer wrap is compromised, the product is no longer considered sterile.

Molds or spreading colonies can make accurate counting difficult.

Unless a statistical method for monitoring is designed, the results can be uninterpretable or misleading.

The complexity of the surface (irregular, curved, porous, rough, or textured) to be tested may present sampling challenges for *contact plates*.

No single assay can characterize completely the microbial contamination in a specific area. A complete contamination control program should emphasize traffic control, special dress code procedures in critical areas, suitable ventilation, as well as good cleaning and disinfecting practices.

PROCEDURE:*

Specimen Collection: Consult appropriate references and texts for the exact method of specimen collection.[2,3]

Method of Use: Prior to inoculation, the medium should be brought to room temperature. For direct specimens, inoculate using aseptic technique, and streak for isolation using the four-quadrant method.

Alternately, specimens may be inoculated using decimal dilutions of the sample, 10^{-1} to 10^{-6}. Inoculate 1.0 ml of sample and 1.0 ml of each dilution onto the medium. Spread the dilution evenly over the surface of the medium. Incubate aerobically/anaerobically for 24-48 hours. Count colonies according to standard guidelines outlined in appropriate references. Medium may be held for up to 7 days. Consult appropriate references for more complete information concerning dilution inoculation.[2,3]

Interpretation: For colony morphology, two possible reactions may be observed:

White to off-white zones surrounding colonies indicate precipitated zones of para-casein. Transparent inner zones surrounding white zones mean digestion of para-caseinate (the proteolytic process has been completed). These reactions indicate the presence of caseolytic microorganisms.

Consult appropriate references for details on enumeration of microorganisms.[2,3]

Material Required but Not Provided: Standard microbiological supplies and equipment such as loops, needles, incubator, and incinerator are not provided.

QUALITY CONTROL:*

Microorganisms Used (ATCC #):	**Expected Results:**
Staphylococcus aureus (25923)	Growth
Escherichia coli (25922)	Growth
Enterococcus faecalis (29121)	Growth
Pseudomonas aeruginosa (27853)	Growth

Key: See “Interpretation”

User Quality Control: Check for signs of contamination and deterioration.

BIBLIOGRAPHY:

1. Bowers and Hucker, *Tech. Bull.#* 228,N.Y. Exp.Station, 1944.
2. Franson, M. A. H. (ed.), *Standard Methods for the Examination of Water and Wastewater*, 20th ed., American Public Health Association, Washington D. C., 1998
3. Wehr, H.M., and Frank, J.F., (eds), *Standard Methods for the Examination of Dairy Products,* 17tj ed. American Public Health Association, Washington, D.C., 2004.

Franson, M. A. H. (ed.), *Standard Methods for the Examination of Water and Wastewater*, 20th ed., American Public Health Association, Washington D. C., 1998

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*For more detailed information, consult appropriate references and/or details in the preface of the PML Technical Manual.

PML MICROBIOLOGICALS, INC.
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