

BD[™] EMB Agar (Eosin Methylene Blue Agar), Modified

INTENDED USE

BD EMB Agar, Modified (formula of Holt-Harris and Teague) is a slightly selective and differential medium for the isolation and differentiation of gram-negative enteric bacilli (*Enterobacteriaceae* and several other gram-negative rods) from clinical specimens.

PRINCIPLES AND EXPLANATION OF THE PROCEDURE

Microbiological method.

EMB Agar is based on a formulation described first by Holt-Harris and Teague in 1916.¹ The original formulation of Holt-Harris and Teague was modified by Levine.² The main difference between the two is the inclusion of sucrose in the Holt-Harris and Teague medium. Sucrose is fermented by certain enterics more readily than lactose.³

BD EMB Agar, Modified contains eosin Y and methylene blue dyes which inhibit grampositive bacteria to a limited degree. The dyes also serve as differential indicators in response to the fermentation of lactose and/or sucrose by micro-organisms. Coliforms produce blueblack colonies whereas *Salmonella* and *Shigella* colonies are colorless or have a transparent amber colour. *Escherichia coli* colonies may show a characteristic green metallic sheen due to the rapid fermentation of lactose.

EMB Agar (either with and without sucrose) is included in the set of low-selectivity isolation media for *Salmonella* from fecal and other specimens.⁴

Gram positive bacteria, such as fecal streptococci, staphylococci and yeasts, may either grow on this medium and form pinpoint colonies, or may be inhibited.

REAGENTS

BD EMB Agar, Modified

Formula* Per Liter Purified Water

Pancreatic Digest of Gelatin	10.0 g
Lactose	5.0
Sucrose	5.0
Dipotassium Phosphate	2.0
Agar	13.5
Eosin Y	0.4
Methylene Blue	0.065

pH 7.2 +/- 0.2

*Adjusted and/or supplemented as required to meet performance criteria.

PRECAUTIONS

IVD . For professional use only. 🛞

Do not use plates if they show evidence of microbial contamination, discoloration, drying, cracking or other signs of deterioration.

Consult **GENERAL INSTRUCTIONS FOR USE** document for aseptic handling procedures, biohazards, and disposal of used product.

STORAGE AND SHELF LIFE

On receipt, store plates in the dark at 2 to 8° C, in their original sleeve wrapping until just prior to use. Avoid freezing and overheating. The plates may be inoculated up to the expiration date (see package label) and incubated for the recommended incubation times.

Plates from opened stacks of 10 plates can be used for one week when stored in a clean area at 2 to 8° C.

USER QUALITY CONTROL

Inoculate representative samples with the following strains (for details, see **GENERAL INSTRUCTIONS FOR USE** document). Incubate plates aerobically at $35 \pm 2^{\circ}$ C for 18 to 24 hours.

Strains	Growth Results
Escherichia coli ATCC™ 25922	Growth good to excellent; blue-black colonies with
	green metallic sheen
Salmonella Typhimurium ATCC 14028	Growth good to excellent; light grey to amber colonies
Shigella flexneri ATCC 12022	Growth fair to good; colorless to light amber colonies
Enterococcus faecalis ATCC 29212	Inhibition partial; colorless colonies
Uninoculated	Purple with a greenish-orange cast, slightly opalescent

PROCEDURE

Materials Provided

BD EMB Agar, Modified (90 mm Stacker™ plates). Microbiologically controlled.

Materials Not Provided

Ancillary culture media, reagents and laboratory equipment as required.

Specimen Types

This product is a selective medium for Gram negative rods that can be used for the isolation of gram-negative enteric bacilli from all types of clinical specimens (see also **PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE**).

Test Procedure

Streak the specimen as soon as possible after it is received in the laboratory. The streak plate is used primarily to isolate pure cultures from specimens containing mixed flora. Alternatively, if material is being cultured directly from a swab, roll the swab over a small area of the surface at the edge; then streak from this inoculated area. A nonselective medium such as Columbia Agar with 5% Sheep Blood must also be streaked to provide an indication of other organisms present in the specimen.

Incubate plates, protected from light, at $35 \pm 2^{\circ}$ C for 18 to 24 h.

Results

Typical colonial morphology is as follows:

Organisms	BD EMB Agar, Modified
E. coli	Large, blue-black, green metallic sheen
Enterobacter/Klebsiella	Large, mucoid, blue-black
Proteus	Large, colorless
Salmonella	Large, colorless to amber
Shigella	Large, colorless to amber
Pseudomonas	Irregular, colorless
Gram-positive bacteria	No growth to slight growth

PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE

On **BD EMB Agar, Modified**, organisms of the family *Enterobacteriaceae* and a variety of other Gram negative rods, such as *Pseudomonas* and *Aeromonas* will grow.³⁻⁵

Gram positive organisms are often not completely inhibited on this medium.

Although certain diagnostic tests may be performed directly on this medium, biochemical and, if indicated, immunological testing using pure cultures is necessary for complete identification. Consult appropriate references. ³⁻⁵

REFERENCES

- 1. Holt-Harris, J.E., and O. Teague. 1916. A new culture medium for the isolation of *Bacillus typhosus* from stools. J. Infect. Dis. 18:596-600.
- 2. Levine, M. 1918. Differentiation of *B. coli* and *B. aerogenes* on a simplified eosin-methylene blue agar. J. Infect. Dis. 23:43-47.
- 3. MacFaddin, J.F. 1985. Media for the isolation cultivation maintenance of medical bacteria. Volume 1. Williams and Wilkins, Baltimore, London.
- Farmer III, J.J. 2003. *Enterobacteriaceae:* introduction and identification. *In:* Murray, P. R., E. J. Baron, J.H. Jorgensen, M. A. Pfaller, and R. H. Yolken (ed.). Manual of clinical microbiology, 8th ed. American Society for Microbiology, Washington, D.C.
- Bopp, C.A., Brenner, F.W., Fields, P.I., Wells, J.G., and N.A. Strockbine. 2003. *Escherichia, Shigella*, and *Salmonella. In:* Murray, P. R., E. J. Baron, J.H. Jorgensen, M. A. Pfaller, and R. H. Yolken (ed.). Manual of clinical microbiology, 8th ed. American Society for Microbiology, Washington, D.C.

PACKAGING/AVAILABILITY

BD EMB Agar, Modified

Cat. No. 254014	Ready-to-use plated media, 20 plates
Cat. No. 254073	Ready-to-use plated media, 120 plates

FURTHER INFORMATION

For further information please contact your local BD representative.

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