Bile Esculin Agar (NCM0117)

Intended Use
Bile Esculin Agar is used for the selective isolation and differentiation of group D streptococci from foods and is not intended for use in the diagnosis of disease or other conditions in humans.

Description
Bile Esculin Agar is based on the formulation described by Swan and further evaluated by Facklam and Moody. Rochaix first noted the value of esculin hydrolysis in the identification of enterococccci. Meyer and Schonfeld added bile to the esculin medium and demonstrated 61 of 62 enterococci strains were able to grow and hydrolyze esculin, while other streptococci could not.

Molecular taxonomic studies of the genus *Streptococcus* have placed enterococci, previously described as group D streptococci, in the genus *Enterococcus*. The ability to hydrolyze esculin in the presence of bile is a characteristic of enterococci and group D streptococci. Swan compared the use of an esculin medium containing 40% bile salts with the Lancefield serological method of grouping, and reported that a positive reaction on the bile esculin medium correlated with a serological group D precipitin reaction. Facklam and Moody found that the bile esculin test provided a reliable means of identifying group D streptococci and differentiating them from non-group D streptococci.

Typical Formulation

Beef Extract	11.0 g/L
Enzymatic Digest of Gelatin	34.5 g/L
Esculin	1.0 g/L
Ox Bile	2.0 g/L
Ferric Ammonium Citrate	0.5 g/L
Agar	15.0 g/L

pH: 6.6 ± 0.2 at 25°C
Formula may be adjusted and/or supplemented as required to meet performance specifications.

Precaution
Refer to SDS

Preparation
1. Suspend 64g 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.
4. Cool 45-50°C.

Test Procedure
Refer to appropriate references for instructions on specific material being tested for group D streptococci.

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

Prepared Appearance: Prepared medium is trace to slightly hazy, opalescent, and gray-yellow.

Expected Cultural Response: Cultural response on Bile Esculin Agar at 35 ± 2°C and examined for growth after 18 - 24 hours incubation.

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Microorganism	Approx. Inoculum (CFU)	Expected Results	
		Recovery	Reaction
Enterobacter aerogenes ATCC® 13048	10-100	≥70%	---
Enterococcus faecalis ATCC® 19433	10-100	≥70%	Black colonies
Enterococcus faecalis ATCC® 29212	10-100	≥70%	Black colonies
Enterococcus faecalis ATCC® 33186	10-100	≥70%	Black colonies
Escherichia coli ATCC® 25922	10-100	≥70%	---
Streptococcus pyogenes ATCC® 19615	> 10^4	Partial to complete inhibition	---
Streptococcus pyogenes ATCC® 12344	> 10^4	Complete inhibition	---

The organisms listed are the minimum that should be used for quality control testing.

Results

Refer to appropriate references and procedures for results.

Expiration

Refer to expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if 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 varying nutritional requirements, some strains may be encountered that grow poorly or fail to grow on this medium.

Storage

Store dehydrated culture media at 2-30°C away from direct sunlight. 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.

References

1. Swan, A. 1954.The use of bile-esculin medium and of Maxted's technique of Lancefield grouping in the identification of enterococci (group D streptococci). J. Clin. Pathol. 7:160.
2. Facklam, R. R., and M. D. Moody. 1970. Presumptive identification of group D streptococci: the bile-esculin test. Appl. Microbiol. 20:245.
3. Rochaix, A. 1924. Milieux a leculine pour le diagnostid differentieldes bacteries du grojps strepto-entero-pneumocoque. Comt. Rend. Soc. Biol. 90:771-772.
4. Meyer, K., and H. Schönfeld. 1926. Über die Unterscheidung des Enterococcus vom Streptococus viridans und die Beziehunger beider zum Strptoccus lactis. Zentralb. Bakteriol Parasitenkd. Infektionskr. Hyg. Abt. I orig. 99:402-416.
5. Schleifer, K. H., and R. Kilpper-Balz. 1987. Molecular and chemotaxonomic approaches to the classification of streptococci, enterococci and lactococci: a review. Syst. Appl. Microbiol. 10:1-19.
6. Vanderzant, C., and D. F. Splittstoesser (eds.). Compendium of methods for the microbiological examination of foods, 4th ed. American Public Health Association, Washington, D.C.
7. www.fda.gov/Food/ScienceResearch/LaboratoryMethods/BacteriologicalAnalytical manualBAM/default.htm.
8. Marshall, R. T. (ed.). 2004. Standard methods for the examination of dairy products, 17th ed. American Public Health Association, Washington, D.C.

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