

## PRODUCT INFORMATION AND QUALITY CONTROL SHEET

# BILE ESCULIN AGAR

### I. INTENDED USE

Bile Esculin Agar is a differential medium for the isolation and presumptive identification of group D streptococci.

### II. SUMMARY AND EXPLANATION

Rochaix<sup>1</sup> first noted the value of esculin hydrolysis in the identification of enterococci. Meyer and Schonfeld<sup>2</sup> added bile to the esculin medium and demonstrated that 61 of 62 enterococci strains were able to grow and hydrolyze esculin, while the other streptococci could not. Swan<sup>3</sup> compared the use of an esculin medium containing 40% bile salts and found that positive reaction on the bile esculin medium correlated with a serological group D precipitin reaction. Facklam and Moody<sup>4</sup> found that the bile esculin test provided a reliable means of identifying group D streptococci and differentiating them from non-group D streptococci.

Lindell and Quinn<sup>5</sup> showed that the medium is also useful in the differentiation of the *Klebsiella-Enterobacter-Serratia* group from other *Enterobacteriaceae*.

### III. PRINCIPLES OF THE PROCEDURE

Organisms positive for esculin hydrolysis hydrolyze the glycoside esculin to esculetin and dextrose. The esculetin reacts with the ferric citrate to form dark brown or black complex. Oxgall (bile) is used to inhibit gram-positive bacteria other than enterococci. Beef Extract and Peptone provide the carbon and nitrogen sources required for growth of a wide variety of organisms, and Agar is the solidifying agent.

### IV. TYPICAL FORMULA AND APPEARANCE

Appearance = greenish to medium amber with bluish cast, slightly opalescent

(Approximate formula\* per liter of processed water)

Beef Extract	3.0 g
Peptone	5.0
Esculin	1.0
Oxgall (Bile)	40.0
Ferric Citrate	0.5
Agar	13.5

\*adjusted and/or supplemented to meet performance criteria.

### V. PRECAUTIONS

This product is for IN VITRO diagnostic use only. Culture specimens may contain microorganisms which can be potentially infectious to the user. Strict adherence to aseptic techniques and established precautions against microbiological hazards should be followed throughout the procedure. Carefully dispose of all items which contact patient specimens or isolated bacteria.

### VI. STORAGE/SHELF LIFE

Media should be stored at 2-8°C (36-46°F). DO NOT FREEZE OR EXPOSE TO HIGH TEMPERATURES. Allow unopened tubes to warm to room temperature prior to inoculation. Prior to and during inoculation procedures, tubes should be handled in a manner that minimizes product exposure to the environment. Product that has exceeded the assigned expiration date noted on the label should not be used.

Do not use tubes that exhibit evidence of drying, cracking, discoloration, microbial contamination or any other signs of deterioration. The presence of excessive condensate may indicate tubes that have been damaged by exposure to temperature extremes.

### VII. SPECIMEN COLLECTION

The quality of culture results depends primarily on the adequacy and condition of the specimen submitted for

examination.

Proper specimen collection techniques must be followed to

ensure the most accurate culture results. Sterile swabs and collection containers should be used. Tubes should be inoculated promptly after specimen collection. If a delay in inoculation is unavoidable, transport medium should be employed. Specimens should be collected prior to the initiation of antimicrobial therapy.

Detailed information on proper specimen collection may be obtained from microbiology reference materials.

### VIII. MATERIALS PROVIDED

Bile Esculin Agar Tubes

### IX. MATERIALS REQUIRED BUT NOT PROVIDED

Incubator maintaining 33-37°C.

Ancillary culture media, reagents and laboratory equipment as required.

### X. PROCEDURE

Inoculate the medium with specimen as soon as possible after it is received in the laboratory and incubate at 35 ± 2°C in an aerobic atmosphere.

### XI. EXPECTED RESULTS

NCCLS CONTROL ORGANISMS (ATCC STRAINS)

*Streptococcus faecalis* (ATCC 29212) Growth with blackening around colonies

*Streptococcus pyogenes* (ATCC 19615) Inhibition (partial to complete) no blackening

### XII. LIMITATIONS

The ability to detect microorganisms by culture techniques can be affected by the following factors: improper specimen collection, storage and inoculation, initiation of anti-infective therapy prior to specimen collection, improper culture incubation temperatures and atmospheres, improper length of culture incubation, and improper storage and handling of culture media.

### XIII. REFERENCES

1. Rochaix. 1924. C.R. Soc. Biol. **90**:771.
2. Meyer, K., and H. Schonfeld. 1926. Uber die Unterscheidung des Enterococcus vom Streptococcus viridans und die Beziehung der beiden zum Streptococcus lactis. Zentralbl. Bakteriol. Parasitenkd. Infektionskr. Hyg. Abt. I Orig. **99**:402-416.
3. 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.
4. Facklam, R.R., and M.D. Moody. 1970. Presumptive identification of group D streptococci: The bile-esculin test. Appl. Microbiol. **20**:245.
5. Lindell, S.S., and P. Quinn. 1975. Use of bile-esculin agar for rapid differentiation of Enterobacteriaceae. J. Clin. Microbiol. **1**:440

### USER QUALITY ASSURANCE/ QUALITY CONTROL PROCEDURES AND INFORMATION

HealthLink recommends that the following quality assurance and quality control procedures be performed on each batch of

product.

### **I. QUALITY ASSURANCE**

The following quality assurance procedures must be performed to assure the product will perform according to its intended use within the assigned expiry date:

1. Daily, document that product storage refrigerator maintains temperature within the recommended range: 2-8°C.
2. Daily, document that laboratory incubator maintains temperature within the recommended range: 22-35°C.

### **II. QUALITY CONTROL**

The following incoming inspection procedures must be performed for each batch (batch = same lot, same shipment) of culture media received in the laboratory:

Inspect plates according to instructions contained in Section VI:  
STORAGE/SHELF LIFE.

Note: Notify Technical Service immediately if media does not meet the inspection criteria.

### **TECHNICAL SERVICE**

HealthLink provides a toll free technical service line (1-800-638-2625) to assist with product usage. To have technical questions answered; please call between the hours of 9:00 am to 5:00 pm EST.

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