BD™ Enterococcosel™ Agar

INTENDED USE

BD Enterococcosel Agar is a selective medium for the isolation and enumeration of fecal streptococci (group D) from clinical specimens.

PRINCIPLES AND EXPLANATION OF THE PROCEDURE

Microbiological method.

This medium is based on the Bile Esculin Agar formulation of Rochaix which was later modified by Isenberg et al. by reducing the bile concentration and by adding sodium azide.[1,2] This modification is supplied as **BD Enterococcosel Agar**. The medium is a standard formulation for the isolation of enterococci.[3-5]

Two peptones provide nutrients. Group D streptococci (including enterococci) hydrolyze esculin to esculetin and glucose. Esculetin reacts with an iron salt to form a dark brown or black complex. Ferric citrate is included as an indicator and reacts with esculetin to produce a brown to black complex. Oxgall is used to inhibit gram-positive bacteria other than enterococci. Sodium azide is inhibitory to gram-negative micro-organisms.[5-7]

REAGENTS

BD Enterococcosel Agar

Formula* Per Liter Purified Water

Pancreatic Digest of Casein	17.0 g
Peptic Digest of Animal Tissue	3.0
Yeast Extract	5.0
Oxgall	10.0
Sodium Chloride	5.0
Esculin	1.0
Ferric Ammonium Citrate	0.5
Sodium Azide	0.25
Sodium Citrate	1.0
Agar	13.5

pH 7.1+/- 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 at 35 ± 2°C in an aerobic atmosphere. Examine the plates after 18 to 24h for amount of growth, colony size, pigmentation and selectivity.

Strains	Growth Results
Escherichia coli ATCC™ 25922	Inhibition partial to complete; colourless colonies
Enterococcus faecalis ATCC 29212	Growth good to excellent; colonies beige, strong black halos
Enterococcus faecium ATCC 19434	Growth good to excellent; colonies beige, strong black halos
Streptococcus pyogenes ATCC 19615	Inhibition (partial to) complete; colourless colonies, no black halos
Uninoculated	Light amber, very light olive-brown hue

PROCEDURE

Materials Provided

BD Enterococcosel Agar (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 differential medium for the isolation of Group D streptococci (including enterococci) 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 and streak from this inoculated area. A nonselective medium such as Columbia Agar with 5% Sheep Blood must also be inoculated to provide an indication of other organisms present in the specimen.
Incubate plates 24 to 48 h at 35 ± 2°C in an aerobic atmosphere.

Results

Typical appearance of the organisms is as follows:

Organisms	BD Enterococcosel Agar
Streptococcus pyogenes (Group A)	No growth to trace growth, no black halos
Streptococcus agalactiae (Group B)	No growth to trace growth, may have black halos
Other streptococci (Non-group D)	No growth to trace growth
Enterococci and *Streptococcus bovis*	Small, translucent with brownish-black to black zones.
Staphylococci	Large, white, opaque
Micrococci	Large, white, grayish
Corynebacteria	Small to large, white to grayish-yellow, smooth and irregular
Candida	Small to large, white
Listeria monocytogenes	Small to large, translucent with brownish-black to black zones
Gram-negative bacteria	No growth to trace growth

PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE

This medium is suitable for the isolation of group D streptococci (*Enterococcus* spp. and *Streptococcus bovis*) from all types of clinical specimens. Consult the references.[6-9]

Although other Gram positive bacteria may grow on the medium, this medium is not recommended for their isolation.
Organisms other than enterococci and others than those mentioned in the **Results** section may be esculin positive and may grow on this medium (e.g. *Pediococcus* and *Lactococcus* species). Therefore, biochemical and serological tests are necessary to confirm the presumptive identification obtained with this medium.

REFERENCES

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3. Meyer, K., and H. Schonfeld. 1926. Über die Unterscheidung des *Enterococcus* vom *Streptococcus viridans* und die Beziehungen beider zum *Streptococcus lactis*. Zentralbl. Bakteriol. Parasitenk. Infektionskr. Hyg. Abt. Orig. 99:402-416.
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6. MacFaddin, J.F. 1980. Biochemical tests for identification of medical bacteria, 2nd ed. Williams & Wilkins, Baltimore.
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9. Chapin, K.C., and T.-L. Lauderdale. 2003. Reagents, stains, and media. *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 Enterococcosel Agar
Cat. No. 254019 Ready-to-use plated media, 20 plates

FURTHER INFORMATION

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