**Reference:** 064-PF0008 **Scharlau Microbiology - Technical Data Sheet**

**Product:** # Bile Esculin Azide Agar

## Specification

Solid medium for the confirmation and enumeration of enterococci in clinical samples and water samples by the membrane filtration method according to ISO 7899-2 and clinical samples.

## Presentation

30 Prepared plates
55 mm Plates for filtration purposes
with: 9 ± 1 ml

**Packaging Details**

1 box containing 3 plastic bags with 3 RD-PACK. Each package contains 10 plates of 55 mm.

| Shelf Life | Storage |
|---|---|
| 6 months | 2-25ºC |

## Composition

Composition (g/l):

| | |
|---|---|
| Tryptone | 17.00 |
| Peptone | 3.00 |
| Yeast extract | 5.00 |
| Bile | 10.00 |
| Sodium chloride | 5.00 |
| Esculin | 1.00 |
| Ammonium ferric citrate | 0.50 |
| Sodium azide | 0.15 |
| Agar | 15.00 |

## Description /Technique

Description:
Bile Esculin Azide Medium is a modification of the classical Bile Esculin proposed by Isenberg, Goldberg and Sampson in 1970, but with a reduction in the amount of bile and the addition of sodium azide. Brodsky and Schieman showed that this medium, also known as Pfizer Enterococci Selective Medium gave the best results using the membrane filtration technique.
The actual formulation according to the ISO Standard 7899-2:2000 is used for the second step in the confirmation and enumeration of enterococci in water by the membrane filtration method. The colonies previously selected in the Slanetz Bartley Agar must be confirmed by a short incubation on Bile Esculin Azide Medium for verification of esculin hydrolysis in a selective environment.
Technique:
After an incubation of 24-48 hours on Slanetz Bartley Agar, the membrane filter showing typical colonies is transferred, with sterile forceps in an upright position, to a pre-warmed plate of Bile Esculin Azide Agar. After two hours of incubation at 44 ± 0.5ºC the membrane filter is inspected. All the typical colonies that show brown to black colour in the surrounding medium are considered positive and therefore intestinal enterococci.
A heterogeneous distribution of the colonies or the presence of abundant and different microorganisms can interfere with the differentiation of positive colonies.

## Quality control

**Physical/Chemical control**

Color : Grey greenish pH: 7.1 ± 0.1 at 25ºC

**Microbiological control**

Membrane Filtration; Practical range 100±20 CFU; Min. 50 CFU (Productivity).
Confrm - Transfer the membrane showing suspicious colonies grown onto other media
Aerobiosis. Incubation at 44 ºC, reading after 2 hours.

| Microorganism | Growth |
|---|---|
| *Enterococcus faecalis ATCC® 19433* | Good - black halo |
| *Enterococcus faecalis ATCC® 29212* | Good - black halo |

**Sterility Control**

Incubation 48 hours at 30-35ºC and 48 hours at 20-25ºC: NO GROWTH
Check at 7 days after incubation in same conditions

## Bibliography

·ATLAS, R.M. & J.W. SNYDER (1995) Handbook of Media for Clinical Microbiology. CRC Press. Boca Raton, Fla, USA.
ALVAREZ, M.V., E. BOQUET, I. de FEZ (1988) Manual de Técnicas en Microbiologia Clínica., AEFA, San Sebastian.
·BRODSKY M.H. & D.A. SCHIEMANN (1976) Evaluation of Pfizer Selective *Enterococcus* and KF media for recovery of fecal streptococci from water by membrane filtration. Appl. Environ. Microbiol. 31 :695-699.
·CHAPIN, K.C. & T-L. LAUDERDALE (2003) Chap. 27: Reagents, Stains and Media : Bacteriology in Manual of Clinical Microbiology 8th edition Volume 1, edited by Murray-Baron-Jorgensen-Pfaller-Yolken. ASM. Washington DC, USA
·FORBES, B.A., D.F. SAHM, & A.S. WEISSFELD (1998) Bailey & Scott's Diagnostic Microbiology 10th edition. Mosby. St. Lous. USA
·ISENBERG, H.D., D. GOLDBERG & J. SAMPSON (1970) Laboratory studies with a Selective *Enterococcus* Medium. Appl. Microbiol. 20:433.
ISENBERG, H. D. (1992) Clinical Microbiology Procedures Handbook, Volume 1. ASM Press, Washington USA.
·ISO Standard 7899-2 (2000) Water Quality. Detection and enumeration of intestinal enterococci. Part 2: Membrane filtration method.
. ISO 11133:2014. Microbiology of food, animal feed and water. Preparation, production, storage and performance testing of culture media.
·MURRAY, P.R., E.J. BARON, J.H. JORGENSEN, M.A. PFALLER & R.H. YOLKEN (2003) Manual of Clinical Microbiology 8th Edition, Volume 1, ASM Press, Washington DC., USA
·McFADDIN, J.F. (1985) Media for Isolation-Cultivation-Identrification-Maintenance of Medical Bacteria. Volume 1. Williams & Wilkins. Baltimore. USA
·PRATT-RIPPIN, K. & M. PEZLO (1992) Identification of Commonly Isolated Aerobic Grampositive Bacteria, in Clinical Microbiology Procedures Handbook, Volume 1 edited by H. D. Isenberg, ASM Press, Washington USA
·RUOFF, K.L., R.A. WHILEY & D.BEIGHTON (2003) Chap. 29: Streptococcus in Manual of Clinical Microbiology 8th edition Volume 1, edited by Murray-Baron-Jorgensen-Pfaller-Yolken. ASM. Washington DC, USA
·WINN, W. Jr., S.ALLEN, W. JANDA, E. KONEMAN, G. PROCOP, P. SCHRECKENBERGER, G. WOODS (2006) Konemasn'sa Color Atlas and Textbook of Diagnostic Microbiology. 6th Edition. Lippincott Williams & Wilkins. Philadcelphia. USA.
. UNE-EN ISO 11133 (2014). Microbiología de los alimentos para consumo humano, alimentación animal y agua.-Preparación, producción, conservación y ensayos de rendimiento de los medios de cultivo.