

TECHNICAL DATA SHEET

Article No. 9423

Bile Esculin Azide Agar, prepared plates

SYNONYMS

Enterococcus Selective Agar

SPECIFICATION

Prepared plates, 90 mm. Solid medium for the confirmation and enumeration of enterococci in water by the membrane filtration method according to ISO 7899-2.

Colour: yellow
pH: 7.1 ± 0.1 at 25 °C

COMPOSITION IN 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

PACKAGING DETAILS

9423-20PLATES

20 prepared plates 90 mm

Content: 21 ± 2 ml

Packaging unit: 1 box with 2 packs of 10 plates/pack. Single cellophane.



GUIDELINES

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 (Art. No. 9134 + 9132) 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 $44 \pm 0.5^\circ\text{C}$ the membrane filter is inspected. All the typical colonies that show brown to black color 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.

After incubation, enumerate all the colonies that have appeared onto the surface of the agar. Typical colonies of *Enterococcus sp.* Show a brown to black colored halo. Each laboratory must evaluate the results according to their specifications. Presumptive isolation of *Enterococcus* must be confirmed with further microbiological and biochemical tests.

MICROBIOLOGICAL CONTROL

Incubate, Membrane filtration w. microorganisms, Slanetz Bartley Agar at 37°C during $44 \pm 4\text{h}$ and transfer in BEA medium.

Aerobic incubation at 44°C , for 2h . Esculine Test.

Microbiological control according to ISO 11133:2014/A1:2018; A2:2020

Microorganism	Growth
<i>Enterococcus faecalis</i> ATCC® 19433, WDCM 00009	Good - Esculin Positive reaction
<i>Escherichia coli</i> ATCC® 25922, WDCM 00013	Inhibited
<i>Enterococcus faecalis</i> ATCC® 29212, WDCM 00087	Good - Esculin Positive reaction
<i>Enterococcus faecium</i> ATCC® 6057, WDCM 00177	Good - Esculin Positive reaction
<i>Aerococcus viridans</i> ATCC® 11563, WDCM 00061	Poor to good - Esculin Negative

Sterility control:

Incubation 48 hours at $30\text{-}35^\circ\text{C}$ and 48 hours at $20\text{-}25^\circ\text{C}$: NO GROWTH.

Check at 7 days after incubation in same conditions.

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STORAGE

2-14 °C

SHELF LIFE

3 months unopened from date of manufacture

Last updated: 07.03.23

