Ethyl Violet Azide Dextrose Agar

M1397

Intended Use:

Recommended for detecting and confirming Streptococci and as confirmative medium for faecal pollution indication in water and other nonclinical or clinical specimens.

Composition**

Ingredients	g / L
Tryptone	20.000
Dextrose (Glucose)	5.000
Dipotassium hydrogen phosphate	2.700
Potassium dihydrogen phosphate	2.700
Sodium chloride	5.000
Sodium azide	0.400
Ethyl violet	0.00083
Agar	15.000
Final pH (at 25°C)	7.0±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 50.8 grams in 1000 ml purified/distilled water.Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.Cool to 45-50°C. Mix well and pour into sterile Petri plates.

Principle And Interpretation

Ethyl Violet Azide Broth is based on the formulation of Litsky et al (1) and is a modification of medium developed by Litsky et al (2) with reduced amount of dextrose and increased dye concentration, making the medium highly specific for Enterococci. The presence of Enterococci acts as a valuable index of faecal or sewage pollution in water (3). Ethyl Violet Azide Dextrose Agar is a modification of Ethyl Violet Azide Broth (M426) where 1.5% agar is added as a solidifying agent. It is used for detection and confirmation of Streptococci. It is based on original formulation of Litsky et al (4). Ethyl Violet Azide Dextrose Agar medium has 0.5% dextrose and was found equally productive as the medium described originally containing 1.5% dextrose. It was found that the medium with the lesser amount of carbohydrate was less adversely affected by heat during sterilization. Litsky et al (1) studied a variety of dyes and selective agents for Streptococci and developed a confirmatory medium using ethyl violet and sodium azide as selective agents. Combination of 0.0083gm% of ethyl violet dye and 0.04gm% of azide provided the best selective action favouring growth of Streptococci. E.V.A. Dextrose Agar contain tryptone as source of carbon, nitrogen, vitamins and minerals. Dextrose is the fermentable carbohydrate. Sodium azide and ethyl violet inhibit gram-positive bacilli and gram-positive cocci other than Enterococci. Dipotassium hydrogen phosphate and potassium dihydrogen phosphate buffer the medium. Sodium chloride provides osmotic balance.

Type of specimen

Clinical samples - faecal samples; Water samples

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (4,5).
For water samples follow appropriate techniques for sample collection, processing as per guidelines and local standards (1).
After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

In Vitro diagnostic use. For professional use only. Read the label before opening the container. Wear protective gloves/ protective clothing/ eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture.Standard precautions as per established guidelines should be followed while handling clinical specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations :

1. Individual organisms differ in their growth requirement and may show variable growth patterns on the medium.

2.Each lot of the medium has been tested for the organisms specified on the COA. It is recommended to users to validate the medium for any specific microorganism other than mentioned in the COA based on the user's unique requirement.
3.Further biochemical and serological tests must be carried out for complete identification.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Light amber coloured, clear to slightly opalescent gel forms in Petri plates

Reaction

Reaction of 5.08% w/v aqueous solution at 25°C. pH : 7.0±0.2

pH

6.80-7.20

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 24-48 hours

Organism	Inoculum (CFU)	Growth	Recovery
Escherichia coli ATCC 25922 (00013*)	>=10^4	inhibited	0%
Enterococcus faecalis ATCC 29212 (00087*)	50-100	good-luxuriant	>50%

Key : (*) Corresponding WDCM numbers.

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (3,4).

Reference

1. Litsky W., Mallmann W. L. and Fifield C. W., 1953, Am. J. Public Health, 43:873.
2. Litsky W., Mallmann W. L. and Fifield C. W., 1955, Am. J. Public Health, 45:104.
3. Greenberg A. E., Trussell R. R. and Clesceri L. S., (Eds.), 1998, Standard Methods for the Examination of Water and Wastewater, 20th Ed., APHA, Washington, D.C.
4. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
6. Lipps WC, Braun-Howland EB, Baxter TE,eds. Standard methods for the Examination of Water and Wastewater, 24th ed. Washington DC:APHA Press; 2023.

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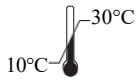

Storage temperature

EC REP

CEpartner4U, Esdoornlaan 13,
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CE Marking

Do not use if package is damaged

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