Azide Dextrose Broth

Intended Use
Azide Dextrose Broth is used for detection and enumeration of Streptococci in water, sewage, food and other materials suspected of sewage contamination.

Summary
Enterococci are more resistant to chlorine in water, hence are better indicators of sewage pollution than *Escherichia coli*. Until 1984, members of the genus *Enterococcus* were classified as Group D Streptococci. Upon genomic DNA analysis, a separate genus status was provided to them. Azide Dextrose Broth is recommended by APHA for enumeration of faecal Streptococci by MPN technique. Azide Dextrose Broth was initially formulated by Rothe, Mullmann and Seligmann for quantitative determination of Enterococci in water, sewage, foods and other materials suspected of contamination with sewage. When large volumes of water samples are to be examined, double strength medium is used. Turbidity in tubes indicates presence of Enterococci, however, it should be further confirmed by inoculation in Ethyl Violet Azide Broth.

Principle
Azide Dextrose Broth (Rothe) is used for the detection of Enterococci in water and sewage. The presence of Enterococci serves as an indicator of faecal contamination. Enterococci are better indicators than *Escherichia coli* of sewage pollution in chlorinated waters because they have a greater resistance to chlorine. Mallmann and Seligmann[2] recommended Azide Dextrose Broth for the quantitative determination of Enterococci in water, sewage, foods and other materials suspected of contamination with sewage.
A blend of peptone and glucose render Azide Dextrose Broth highly nutritious, and sodium chloride maintains osmotic equilibrium. The use of sodium azide as an inhibitor of Gram-negative organisms has been reported by several workers, and the concentration selected provides optimum protection for the Enterococci while largely suppressing the Gram-negative flora. The phosphate buffer system controls pH.

Formula*
Ingredients	**g/L**
Peptone special	15.0
Beef Extract	4.5
Dextrose	7.5
Sodium Chloride	7.5
Sodium Azide	0.2
Final pH (at 25°C)	7.2 ± 0.2

*Adjusted to suit performance parameters

Storage and Stability
Store dehydrated medium below 30°C in tightly closed container and the prepared medium at 2°C-8°C. Avoid freezing and overheating. Use before expiry date on the label. Once opened keep powdered medium closed to avoid hydration.

Type of Specimen
Food samples; Water and sewage samples

Specimen Collection and Handling
Ensure that all samples are properly labelled.
Follow appropriate techniques for handling samples as per established guidelines.
Some samples may require special handling, such as immediate refrigeration or protection from light, follow the standard procedure The samples must be stored and tested within the permissible time duration.
After use, contaminated materials must be sterilized by autoclaving before discarding.

Directions

1. Suspend 34.70 g of the powder in 1000 mL purified / distilled water.
2. Mix thoroughly.
3. Boil with frequent agitation to dissolve the powder completely.
4. Dispense into tubes.
5. Sterilize by autoclaving at 118°C (12 psi) for 15 minutes as per validated cycle.

Quality Control

Dehydrated Appearance: Cream to yellow coloured, homogeneous, free flowing powder.
Prepared Appearance: Light amber to amber coloured, clear solution without any precipitate.
Growth Promotion Test: Growth promotion is carried out in accordance with the harmonized method of USP/EP/JP/IP and growth is observed after an incubation at 30°C-35°C for 18-24 hours.
Growth Promoting Properties: The test results observed are within the specified temperature and shortest period of time specified in the test, inoculating ≤ 100 cfu of appropriate microorganism at 30°C-35°C for 18 hours.
Inhibitory Properties: No growth of the test microorganism occurs for the specified temperature and longest period of time, inoculating > 100 cfu at 30°C-35°C for ≥ 24 hours.

Organism (ATCC)	**Growth**
Enterococcus faecalis (29212)	Good
Escherichia coli (25922)	Inhibited

Note:

1. No growth of the organism should occur for the inhibitory test.
2. Inoculum for Good growth is 10 – 100 cfu and that for inhibition is greater than 100 cfu.

Performance and Evaluation

Performance of the product is dependent on following parameters as per product label claim:

1. Directions
2. Storage
3. Expiry

Precaution

In vitro diagnostic 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.

Warranty

This product is designed to perform as described on the label and package insert. The manufacturer disclaims any implied warranty of use and sale for any other purpose.

Reference

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16. Data on file: Microxpress®, A Division of Tulip Diagnostics (P) Ltd.

Product Presentation:

Cat No.	Product description	Pack Size
201010340500	Dehydrated Culture Media	500 g

Disclaimer
Information provided is based on our inhouse technical data on file, it is recommended that user should validate at his end for suitable use of the product.