Azide Dextrose Broth

M345

Intended use

Recommended is used for detection and enumeration of Streptococci in water, sewage, food and other materials suspected of sewage contamination.

Composition**

Ingredients	Gms / Litre
Peptone, special	15.000
HM peptone B #	4.500
Dextrose (Glucose)	7.500
Sodium chloride	7.500
Sodium azide	0.200
Final pH (at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters
Equivalent to Beef extract

Directions

Suspend 34.7 grams in 1000 ml purified/ distilled water for preparing single strength broth or use 69.4 grams in 1000 ml purified / distilled water for double strength broth. Heat, if necessary, to ensure complete solution. Dispense in test tubes and sterilize by autoclaving at 118°C for 15 minutes.

Principle And Interpretation

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. (12). 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 (9,10) 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 (M426).

Azide Dextrose Broth is a highly nutritious medium due to the presence of nutrient rich peptone special, HM peptone B and dextrose. Sodium azide inhibits growth of gram-negative bacteria, allowing Enterococci to grow (3,4,5). Streptococci detected by the above media should be further identified using chemicals (8).

Type of specimen

Food samples; Water and sewage samples

Specimen Collection and Handling:

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (1,11,13).
For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards.(2)
After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

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 specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations :

1. Further biochemical tests must be carried out for confirmation.

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

Colour and Clarity of prepared medium

Amber coloured clear solution without any precipitate.

Reaction

Reaction of 3.47% w/v aqueous solution at 25°C. pH : 7.2±0.2

pH

7.00-7.40

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours

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

Key : *Corresponding WDCM numbers.

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°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. Use before expiry date on the label.
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 (6,7).

Reference

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