

Glucose Azide Broth**M982**

Glucose Azide Broth is used for the enumeration of faecal Streptococci by MPN technique from water and sewage.

Composition**

Ingredients	Gms / Litre
Peptic digest of animal tissue	10.000
Yeast extract	3.000
Sodium chloride	5.000
Dipotassium phosphate	5.000
Monopotassium phosphate	2.000
Dextrose	5.000
Sodium azide	0.250
Bromo cresol purple	0.030
Final pH (at 25°C)	6.7±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 30.28 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Dispense 5 ml amounts in 16 x150 mm test tubes and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. For large inocula of 5 ml or more quantities, prepare double strength medium.

Warning: Sodium azide has a tendency to form explosive metal azides with plumbing materials. It is advisable to use enough water to flush off the disposables .

Principle And Interpretation

Faecal *Streptococcus* is a group of bacteria normally present in large numbers in the intestinal tract of warm-blooded animals other than human. The faecal Streptococcus group consists of a number of species of the genus Streptococcus such as *Streptococcus faecalis*, *Streptococcus faecium*, *Streptococcus avium*, *Streptococcus bovis*, *Streptococcus equines* and others. They have been used with faecal coliforms to differentiate faecal contamination from humans and from that of other warm-blooded animals. They tend to persist longer in the environment than faecal coliforms. Glucose Azide Broth is recommended by Hannay and Norton (1) for enumeration of faecal streptococci by MPN technique from water, sewage, foods and other materials suspected to be contaminated with sewage.

Glucose Azide Broth is a highly nutritious medium due to its content of peptic digest of animal tissue, yeast extract and dextrose, which provide nitrogenous compounds, carbon, sulphur, amino acids and trace ingredients. Sodium chloride maintains osmotic balance of the medium. Sodium azide suppresses the growth of gram-negative organisms and thereby allows the selective growth of faecal Streptococci.

Prior to inoculation, warm the tubes of Glucose Azide Broth to 44-45°C by heating in a water bath. Inoculate the tubes containing Glucose Azide Broth with heavy inocula from all the positive presumptive test tubes. MacConkey Broth purple (M083) is generally used for the presumptive test. Incubate inoculated Glucose Azide Broth tubes at 44-45°C for 24-48 hours. Tubes showing yellow colour change, due to acid production are subcultured onto Bile Esculin Azide Agar (M493) for confirming the presence of faecal streptococci (2).

Quality Control**Appearance**

Light yellow to beige homogeneous free flowing powder

Colour and Clarity of prepared medium

Purple coloured, clear solution without any precipitate

Reaction

Reaction of 3.03% w/v aqueous solution at 25°C. pH : 6.7±0.2

Cultural Response

M982: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Colour change to yellow			
<i>Enterococcus faecalis</i> ATCC 29212	50-100	good-luxuriant	positive			

<i>Enterococcus hirae</i> ATCC 8043	50-100	good-luxuriant	positive			
<i>Staphylococcus aureus</i> ATCC 25923	$\geq 10^3$	inhibited	negative			
<i>Escherichia coli</i> ATCC 25922	$\geq 10^3$	inhibited	negative			

Reference

1. Hannay C. L., Norton I. L., 1947, Proc. Soc. Appl. Bacteriol. 1: 59
2. Collee J. G., Duguid J. P., Fraser A. G., Marmion B. P., (Eds) Mackie and McCartney, Practical Medical Microbiology, 1989, 13th Edition, Churchill Livingstone

Storage and Shelf Life

Store below 30°C and prepared medium at 2-8°C. Use before expiry period on the label.