

Azide Dextrose Broth

Used as a preliminary test for enterococci and also for their selective enrichment.

See also Bromocresol-purple Azide Broth.

Mode of Action

The concentration of sodium azide present in this medium largely inhibits the growth of the accompanying Gram-negative microbial flora, while sparing the enterococci.

The use of sodium azide as a selective inhibitor for Gram-negative bacteria was reported in the studies of EDWARDS (1933, 1938) and HARTMANN (1936) on the isolation of *Str. agalactiae*. MALLMANN (1940) and SNYDER and LICHSTEIN (1940) later showed that sodium azide can also be used for the isolation of enterococci from water.

The presence of enterococci (*Enterococcus faecalis*, *S. durans*, *S. bovis* and *S. equinus*) serves as an indicator for faecal contamination, particularly when this took place a long time ago and the less resistant coliform bacteria, including *E. coli*, may be already dead when the analysis is carried out.

Typical Composition (g/litre)

Peptone from casein 15.0; meat extract 4.5; D(+)-glucose 7.5; sodium chloride 7.5; sodium azide 0.2.

Preparation

Suspend 35 g or 70 g/litre, dispense into suitable vessels, autoclave (15 min at 121 °C). **Do not overheat.**

pH: 7.2 ± 0.2 at 25 °C.

The prepared broth is clear and yellowish-brown.

Experimental Procedure and Evaluation

Small sample volumes (up to 1 ml) can be added to the normal strength broth. Larger volumes (10 ml or more) should be diluted with an equal volume of the double-strength broth.

Incubation 24-48 hours at 35 °C aerobically.

If the broth becomes turbid due to microbial growth it is likely that enterococci are present. The culture should then be inoculated into Bromocresol-purple Azide Broth. If this broth does not become turbid enterococci are not present.

Literature

American Public Health Association, American Water Works Association and Water Pollution Control Federation: Standard Methods for the Examination of Water and Wastewater, 20th ed., Washington, 1998.

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LITSKY, W., MALLMANN, W.L., a. FIFIELD, C.W.: A new medium for the detection of enterococci in water. - *Amer. J. Publ. Hlth.*, **43**: 873-879 (1953).

HARTMANN, G.: Ein Beitrag zur Reinzüchtung von Mastitisstreptokokken aus verunreinigtem Material. - *Milchw. Forsch.*, **18**: 116-122 (1936).

MALLMANN, W.L.: A new yardstick for measuring sewage pollution. - *Sewage Works J.*, **12**: 875-878 (1940).

SNYDER, M.L., a. LICHSTEIN, H.C.: Sodium azide as an inhibiting substance for Gram-negative bacteria. - *J. Infect. Dis.*, **67**: 113-115 (1940).

Verordnung über Trinkwasser und über Wasser für Lebensmittelbetriebe (Trinkwasserverordnung) vom 22. Mai 1986. - *Bundesgesetzblatt*, Teil I, 760-773 (1986).

Ordering Information

Product	Merck Cat. No.	Pack size
Azide Dextrose Broth	1.01590.0500	500 g
Bromocresol-purple Azide Broth	1.03032.0500	500 g



100 ml sample into 100 ml of double-strength Azide Dextrose Broth

Quality control

Test strains	Growth
<i>Enterococcus faecalis</i> ATCC 11700	good / very good
<i>Enterococcus faecalis</i> ATCC 19433	good / very good
<i>Enterococcus hirae</i> ATCC 8043	good / very good
<i>Streptococcus bovis</i> DSMZ 20065	fair / very good
<i>Staphylococcus aureus</i> ATCC 25923	none / poor
<i>Escherichia coli</i> ATCC 25922	none / poor
<i>Pseudomonas aeruginosa</i> ATCC 27853	none / poor