Dehydrated Culture Media

SLANETZ AND BARTLEY

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MEDIUM

Code: CM0377

A medium for the detection of enterococci.

Typical Formula*

	gm/litre
Tryptose	20.0
Yeast extract	5.0
Glucose	2.0
Di-potassium hydrogen phosphate	4.0
Sodium azide	0.4
Tetrazolium chloride	0.1
Agar	10.0

pH 7.2 ± 0.2 @ 25°C

*** Adjusted as required to meet performance standards**

Directions
Suspend 42g in 1 litre of distilled water and bring to the boil to dissolve the agar completely.

EXCESSIVE HEATING MUST BE AVOIDED. Dispense into Petri dishes and allow to solidify. It should not be remelted. The medium may be used with membrane filters or by spreading dilutions of the sample over the surface of the agar with a glass rod.

Description

Slanetz & Bartley[1] originally devised this medium to detect and enumerate *enterococci* by the technique of membrane filtration, but it has also proved useful as a direct plating medium[2,3].

The medium is very selective for enterococci and, when it is incubated at elevated temperatures (44-45°C), all red or maroon colonies may be accepted as presumptive *enterococci*[4,5].

Burkwall and Hartman showed that the addition of 0.5ml of `Tween 80' and 20ml of a 10% solution of sodium carbonate or bicarbonate to each litre of medium of a modified formulation of Slanetz and Bartley Medium was of value when examining frozen foods for enterococci; the original article should be consulted for procedural details [2].

Technique

The Environment Agency 'Microbiology of Drinking Water 2002'[6] recommend the use of Slanetz and Bartley medium for the enumeration of *enterococci* in water supplies, as do ISO in the standard for water quality[7]. The water is filtered through a membrane filter which is then placed on the surface of a well dried plate of the medium. Plates are incubated at 35°C for 4 hours and then at 44-45°C for 44 hours. Membranes are examined, with a hand lens in a good light, and all red or maroon colonies counted as *enterococci.*

Food samples can be examined for enterococci by the method suggested by the Nordic Committee of Food Analysis[3]. Samples are homogenised and so diluted with physiological saline that only 15-150 colonies grow on each Petri dish. Homogenates or dilutions are spread evenly over the agar surface with a glass rod and allowed to soak in. Dishes are inverted and incubated at 35°C for 48 hours, after which typical colonies (pink or dark red, with a narrow whitish border) are counted.

Storage conditions and Shelf life
Store the dehydrated medium at 10-30°C and use before the expiry date on the label.
Store the prepared medium at 2-8°C away from light.

Appearance
Dehydrated medium: Straw coloured, free-flowing powder
Prepared medium: Straw coloured gel

Quality control

Positive control:
Expected results
Enterococcus faecalis ATCC® 29212 *
Good growth; deep red coloured colonies
Negative control:

Escherichia coli ATCC® 25922 *
No growth

*** This organism is available as a Culti-Loop®**

Precautions
Count all red, maroon or pink colonies as presumptive enterococci. Not all species reduce TTC therefore pale colonies should not be ignored.
Although incubation at 35°C yields a higher count, it allows the growth of organisms which do not conform to the definition of enterococci. Incubation at 44-45°C has a selective effect and produces fewer false-positives. However, the preliminary incubation at 35°C encourages the recovery of stressed organisms.
Although the selective properties of this medium are very good it is advisable to regard the colony count as a presumptive or unconfirmed count. Further identification may be required depending on the scope of the examination.

References
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5. Mead G. C. (1966) *Proc. Soc. Wat. Treat. Exam*. 15. 207-221.
6. Environment Agency '*Microbiology of Drinking Water 2002'. Methods for Examination of Waters and Associated Materials.*
7. ISO Standard for Water Quality - Detection and enumeration of intestinal enterococci - Part 2 : Membrane filtration method.

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