



CHROMOGENIC E. COLI AGAR (CHROMOGENIC TRYPTONE BILE GLUCURONIDE AGAR) (TBX AGAR) (ISO) TM 1339

INTENDED USE

For easy enumeration of *E. coli* without membrane filtration, or pre-incubation on Mineral Modified Glutamate Medium Base

COMPOSITION

Ingredients	Gms/Ltr
Peptone	20.000
Agar	15.000
Bile salts	1.500
X-β-D-Glucuronide	0.075

PRODUCT SUMMARY AND EXPLANATION

TBX Agar is based on Tryptone Bile Agar. Tryptone Bile Agar was originally formulated to improve on earlier methods used to detect *Escherichia coli* in foods in terms of speed, reliability, better recovery from frozen samples and the detection of poor lactose fermenters.

TBX medium builds on these advantages through the addition of a chromogenic agent - X-glucuronide - which detects glucuronidase activity. This is the same enzyme detected by MUG reagent, and has been shown to be highly specific for *E. coli*. However, approximately 3-4% of *E. coli* are glucuronidase negative, notably *E. coli* O157 strains.

Most *E. coli* strains can be differentiated from other coliforms by the presence of the enzyme glucuronidase. The chromogen in TBX Agar is 5-bromo-4-chloro-3-indolyl-beta-D-glucuronide (X-glucuronide), and is targeted by this enzyme. *E. coli* cells are able to absorb this complex intact and intracellular glucuronidase splits the bond between the chromophore and the glucuronide. The released chromophore is coloured and builds up within the cells, causing *E. coli* colonies to be coloured blue/green.

PRINCIPLE

The chromogenic agent X- Glucuronide used in this medium helps to detect glucuronidase activity. *Escherichia coli* cells absorb x-glucuronide and the intracellular glucuronidase splits the bond between the chromophore and the glucuronide. The released chromophore gives coloration to the colonies. The peptone provides the essential growth nutrients to the organisms. Bile salts mixture inhibits gram-positive organisms.



INSTRUCTION FOR USE

1. Dissolve 36.57 g in 1000 ml distilled water.
2. Gently heat in a boiling water bath or in flowing steam until the medium is completely dissolved.
3. Autoclave at 15 psi (121°C) for 15 min.
4. Cool to 45-50°C in a water bath, mix gently and pour into sterile petri dishes.

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder: Cream to yellow colour, homogenous mixture, free flowing powder

Appearance of prepared medium: Light yellow colour, clear to slightly opalescent gel

pH (at 25°C): 7.2 ± 0.2

INTERPRETATION:

Culture characteristics observed after incubation period of 18 - 24 hours at 44°C.

Microorganisms	ATCC	Inoculum (CFU)	Appearance of colony	Standard recovery (%)
<i>Escherichia coli</i>	25922	50-100	Bluish green	≥ 50%
<i>Salmonella enteritidis</i>	13076	50-100	Colourless	≥ 50%
<i>Staphylococcus aureus</i>	25923	≥ 1000	Inhibited	0%

STORAGE & STABILITY

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers below 8°C and protect from direct Sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

REFERENCES

1. Gross R.J. and Rowe B. (1985) J. Hyg. Lond. 95. 531-550.
2. Anderson J.M. and Baird-Parker A.C. (1975) J. Appl. Bact. 39. 111-117.
3. Feng P.C.S. and Hartman P.A. (1982) Appl. Environ. Microbiol. 43. 1320-1329.
4. Hansen W. and Yourassowsky E. (1984) J. Clin. Microbiol. 20. 1177-1179.
5. Ratnam S., March S.B., Almed R., Bezanson G.S. and Kasatiya S. (1988) J. Clin. Microbiol. 26. 2006-2012.



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PRODUCT DATA SHEET



NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.