Technical Data Sheet

ReadyPlate™ CHROM TBX (Tryptone Bile X-glucoronide) Agar

Ordering number: 1.46326.0020

For the enumeration, detection and identification of β-glucuronidase positive Escherichia coli from food and feeding stuff as well as from environmental samples in the area of food production and food handling.

General

This culture medium complies with the specifications given by ISO 16649 part 1 and part 2 and with EN ISO 16649-1..

Mode of Action

The presence of the enzyme β-D-glucuronidase differentiates most Escherichia coli from other coliforms. E. coli absorbs the chromogenic substrate 5-bromo-4-chloro-3-indolyl-β-D-glucuronide (X-β-D-glucuronide). The enzyme β-glucuronidase splits the bond between the chromophore 5-bromo-4-chloro-3-indolyle- and the β-D-glucuronide. E. coli colonies are then coloroud blue to blue-green.

Some E. coli (3-4%) are β-D-glucuronidase-negative and appear as colourless colonies, e.g. most E. coli O157 strains or they cannot grow at the elevated temperature of 44 °C, e.g. E. coli 0157:H7. For the detection of E. coli O157 specific culture media have to be used.

Growth of accompanying Gram-positive flora is largely inhibited by the use of bile salts and the high incubation temperature of 44 °C.

Typical Composition (g/l)

Specified by ISO 16649		ReadyPlate™ CHROM TBX	
Enzymatic digest of casein	20	Enzymatic digest of casein	20
Bile salts No. 3	1.5	Bile salts No. 3	1.5
5-Bromo-4-chloro-3-indolyl-ß-D-glucuronic acid (BCIG)	144 µmol[a]	5-Bromo-4-chloro-3-indolyl-β-D-glucuronic acid (BCIG) cyclohexylammonium salt [a]	0.075
Agar	9 to 18	Agar	16
Water	1000	Water	1000
pH at 25°C	7.2±0.2	pH at 25°C	7.2±0.2
[a] ISO 16649 specifies: 5-Bromo-4-chloro-3-indolyl-ß-D-glucuronic acid (BCIG) 144 µmol - for example 0.075 g of cyclohexylammonium salt.			

The appearance of the medium is clear and yellowish.

Application and Interpretation

Please check each agar plate before using it on sterility and pay attention to aseptic handling in order to avoid false positive results.

Depend on the purpose for which the medium is used.

Incubate the inoculated plates under aerobic conditions. e.g. acc. to ISO 16649-1/-2 und EN ISO 16649-1 at 44 ± 1 °C for 22 h ± 2 h.

The presence of blue or blue green colonies indicates the presence of β-glucuronidase positive E. coli.

If the presence of stressed cells is suspected, incubate for an initial period of 4 h at 37 °C, and then raise the incubation temperature to 44 °C for 18 h to 24 h. The incubation temperature shall not exceed 45 °C

Storage and Shelf Life

The product can be used for sampling until the expiry date if stored upright, protected from light and properly sealed at 2 to 8°C.

Condensation can be prevented by avoiding quick temperature shifts and mechanical stress.

The testing procedures as described on the CoA can be started up to the expiry date printed on the label.

Disposal

Please mind the respective regulations for the disposal of used culture medium (e.g. autoclave for 20 min at 121 °C, disinfect, incinerate etc.).

Quality Control

Function	Incubation	Control strains	Method of control	Criteria	Characteristic reactions
Productivity Selectivity	(21±3) h / (44±1)°C	*Escherichia coli* ATCC® 25922 (WDCM 00013)	Qualitative	≥ 50%	Turquoise to blue colonies
		Escherichia coli NCTC 13216 (WDCM 00202)			
Selectivity	(21±3) h / (44±1)°C	*Enterococcus faecalis* ATCC® 19433 (WDCM 00009)		Total inhibition	-
		Enterococcus faecalis ATCC® 29212 (WDCM 00087)			
Specificity	(21±3) h / (44±1)°C	*Citrobacter freundii* ATCC® 43864 (WDCM 00006)	Qualitative	-	White to green-beige colonies
		Pseudomonas aeruginosa ATCC® 27853 (WDCM 00025)			

A recovery rate of 50 % is equivalent to a productivity value of 0.5.

Literature

ISO International Standardisation Organisation. Microbiology of food and animal feeding stuffs - Horizontal method for the detection of *Salmonella spp.* EN ISO 6579:2002.

ISO 16649-1-3: Horizontal method for the enumeration of beta-glucuronidase-positive Escherichia coli.

Part 1 (2001): Colony-count technique at 44 °C using membranes and 5-bromo-4-chloro-3-indolyl beta-D-glucoronide.

Part 2 (2001): Colony-count technique at 44 °C using 5-bromo-4-chloro-3-indolyl beta-D-glucuronide. Part 3 (2015): Most probable number technique using 5-bromo-4-chloro-3-indolyl-beta-D-glucuronide.

Ratnam S, March S. B., Ahmed R., Bezanson G. S., Kasatiya S. (1988): Characterization of Escherichia coli Serotype O157:H7. J. Clin. Microbiol. 26: 2006-2012.

ISO International Standardisation Organisation. Microbiology of the food chain

Ordering Information

Product	Article number	Pack size
ReadyPlate™CHROM TBX Agar ISO 16649	1.46326.0020	20 x 90mm
Chromocult® TBX Agar ISO 16649	1.16122.0500	500g
Granucult MMGA (Mineral Modified Glutamate) Agar	1.09045.0500	500g

Lit.No. MK_PF11855EN