

Art. 84641.0500

Technical Data Sheet

TCBS AGAR

DEHYDRATED MEDIUM

Also known as

Cholera Medium TCBS

Intended use

Solid medium for the selective isolation of Vibrio spp, and Vibrio parahaemolyticus according to the ISO standard.

Formula * - Composition in q/L

Proteose peptone	10.000	Ferric citrate	1.000	
Yeast extract		Thymol blue	0.040	
Sodium citrate		Bromthymol blue	0.040	
Sodium thiosulphate		Agar	14.000	
Ox bile	8.000			
Sucrose	20.000	Final pH 8.6 ±0.2 at 25 °C		
Sodium chloride	10.000			

^{*} Adjusted and /or supplemented as required to meet performance criteria

Instructions for preparation

Suspend 88 g of powder in 1 l of purified water. Heat with constant stirring until boiling. Pour immediately into plates. Do not sterilise and avoid remelting.

Principle of the method and general information

TCBS Agar is universally accepted as the medium of choice for differential isolation of enteropathogenic vibrios, whilst inhibiting all the accompanying organisms. This formulation provides high growth of *Vibro cholerae* and *V. parahaemolyticus*. *V. alginoliticus* and NAG-vibrios. Enterobacteria are strongly inhibited by high concentrations of citrate, thiosulfate, bile and sodium chloride.

Although some enteric bacteria may also grow in this medium, their colony morphology is quite different to that of *Vibrio spp*.

The organisms that can be confused with vibrios are some biotypes of *Proteus* and *Pseudomonas*. There are some resistant enterococci which may form exceptionally small and yellow colonies on this medium. Usually, colonies are selected or chosen and then identified with primary tests [oxidase reactions in Kligler Iron Agar, MRVP Broth, and antibiotic sensitivity test] before performing serological identification and phage typing.

Due to its high selectivity, the medium can be seeded with large inoculum of pathological material. Once solidified and cooled, the medium is turbid, but the observations are not affected.

This medium is very thermolabile and so it must not be autoclaved, overheated or re-melted.

Colonial appearance on TCBS Agar after 24 hours at 37°C:

- Vibrio alginolyticus and Vibrio cholerae: Large, yellow colonies.
- Vibrio parahaemolyticus: Small, yellow, without halo and with a green core.
- Streptococcus faecalis: Very small and convex, yellow with yellow halo.
- Enterobacteria generally: Small and transparent.
- Pseudomonas, Aeromonas, Proteus: meaf medium size and blue.
- Some strains of *Vibrio cholerae* and *Vibrio parahaemolyticus* carry out delayed sucrose fermentation so they produce medium sized colonies, and are colourless or dirty yellow with a dark nucleus.

Quality control

Incubation temperature: 37°C ±1.0 Incubation time: 24±3 h

Inoculum: Previous enrichment. 6±1h (ASPW). Streak isolation. (ISO 11133:2014/Amd 1:2018)

Microorganism	Growth	Remarks

Vibrio parahaemolyticus ATCC® 17802GoodBlue-green colonies 3-5mmØVibrio algynoliticus ATCC® 17749GoodYellow colonies 3-5mmØVibrio furnissii NCTC® 11218GoodYellow colonies 3-5mmØEscherichia coli ATCC® 8739Inhibited-





Technical Data Sheet

References Art. 84641.0500

- · ATLAS, R.M. & L.C. PARKS (1993) Handbook of Microbiological Media CRC Press. BocaRaton. Fla. USA
- · BHATTACHARYA, M.K., S.K. BATTACHARYA, S. GARG, P.K. SAHA, D. DUTTA, G.B. NAIR, B.C. DEB & K.P. DAS (1993) Outbrek of Vibrio cholerae non-01 in India and Bangladesh. Lancet, 341:1345-1347.
- DOWNES, F.P. & K. ITO (2001). Compendium of Methods for the Microbiological Examination of Foods. 4th ed. APHA. Washington. DC. USA.
- · FORBES, B.A., D.F SAHM & A.S. WEISSFELD (Eds) (1998) Bailey & Scott's Diagnostic Microbiology 10th ed. Mosby. St. Louis, MO. USA.
- · HORWITZ, W. (Ed) (2000) Official Methods of Análisis of AOAC Internacional. 17th ed. Gaithersburg. MD. USA.
- · ISO 21872-1 Technical Specification (2017) Microbiology of Food chain- Horizontal method for the detection of potentially enteropathogenic Vibrio spp. Part 1: Detection of *Vibrio parahaemolyticus and Vibrio cholerae and Vibrio vulnificus*.
- . ISO 11133:2014/ Adm 1:2018. Microbiology of food, animal feed and water. Preparation, production, storage and performance testing of culture media.
- · KOBAYASHI, T., ENOMOTO, S. SAKAZARI, R. and KUWAHARA, S. (1963) A new selective medium for patogenic vibrios: TCBS (modified Nakanishi Agar) Jap. J.Bact. 18:387.
- MacFADDIN, J.F. (1985) Media for isolation-cultivation-identification-maintenance of medical bacteria. Williams & Wilkins. Baltimore. MD. USA.
- MURRAY, P.R., E.J. BARON, J.H. JORGENSEN, M.A. PFALLER & R.H. YOLKEN (Eds) (2003) Manual of Clinical Mcrobiology 8th ed. ASM Press. Washington. DC. USA.
- · PASCUAL ANDÉRSON, MªRª (1992) Microbiología Alimentaria. Díaz de Santos, S.A. Madrid.
- · US FDA (Food and Drug Administrations) (1998) Bacteriological Analytical Manual 8th ed. Revision A. AOAC International Inc. Gaithersburg. MD. USA.

Storage conditions

For laboratory use only. Keep tightly closed, away from bright light, in a cool dry place (+4 °C to 30 °C).

Ordering information

84641.0500 TCBS AGAR Bulk of 500 g.

Note: For supplements see the section - Instructions for preparation.

