Thiosulfate Citrate Bile Salts Sucrose (TCBS) Agar Plate (M920)

This medium is recommended for selective isolation of Vibrio cholerae and other enteropathogenic Vibrio species.

Composition

Ingredients	g/ L
Peptone	10.000
Yeast extract	5.000
Sodium citrate	10.000
Sodium thiosulphate	10.000
Sodium cholate	3.000
Bile	5.000
Sucrose	20.000
Sodium chloride	10.000
Ferric citrate	1.000
Bromo thymol blue	0.040
Thymol blue	0.040
Agar	15.000

Description

TCBS Agar is a selective medium widely used to isolate and cultivate practically all bacteria of the genus Vibrio, including V. cholerae and V.alginolyticus, pathogenic to humans causing cholera, choleral diarrhea or food poisoning from contaminated foods and from stool specimens. The last 2 conditions especially can be caused by ingesting raw or partially processed fish or seafood containing Vibrio parahemolyticus. The only Vibrio that does not grow in TCBS is V. hollisae.

The meat and casein peptones provide nitrogen, vitamins, minerals and amino acids essential for growth. Yeast extract is a source of vitamins, particularly of the B-group. Sodium citrate, sodium thiosulfate and ox bile are the selective agents, inhibiting the Gram positive bacteria. Sodium thiosulfate provides sulphur, and ferric citrate is the indicator for H2S production. Sucrose is the carbohydrate energy source. Bromothymol blue and thymol blue are pH indicators. Sodium chloride promotes growth (Vibrio grows well in salty media). Bacteriological agar is the solidifying agent. The alkaline pH of the medium enhances the recovery of V. cholerae.

Sucrose-positive vibrios, such as Vibrio cholerae and Vibrio alginolyticus, are yellow on TCBS. Sucrose-negative ones, such as Vibrio para-haemolyticus and Vibrio vulnificus, produce blue- green colonies. Almost all Vibrio ferment sucrose and yield yellow colonies from the production of acid. Some types of Proteus (fermenters of sucrose) can form yellow colonies similar to those of Vibrio.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines. After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

In Vitro diagnostic use only. For professional use only. Follow good microbiological lab practices while handling specimens and culture.

Limitations

1. Individual organisms differ in their growth requirement and may show variable growth patterns.
2. Each lot of the medium has been tested for the organisms specified on the COA.
3. The surface of the medium should be dry when inoculated.
4. Occasional isolates of Pseudomonas and Aeromonas may also form blue green colonies on TCBS Agar.
5. Proteus species that are sucrose-fermenters may form yellow colonies.
6. TCBS Agar is not a suitable medium for oxidase testing of Vibrio species.
7. A few strains of V.cholerae may appear green or colourless on TCBS Agar due to delayed sucrose fermentation.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality control

Physical/Chemical control

Color : Bluish green pH: 8.60-9.00

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours.

Microbiological activity

Organisms	Inoculum CFU/ml	Growth
Escherichia coli ATCC 25922	>=10^3	Inhibited
Vibrio cholerae ATCC 15748	50-100	Yellow colonies
Vibrio parahaemolyticus ATCC 17802	50-100	Bluish green colonies

Storage and Shelf Life

Store between 2-8°C. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques.

Reference

1. Cholera Information (WHO, 1965). WHO Expert Committee on Cholera (2 and Rep. Techn., Rep. Series No. 352. 1967. Felsemfeld, Bull.
2. World Otg. 34:161. 1966. Kobayashi. T. Enomoto S. Sakasaki, R. Y.
3. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Yolken R. H., (Eds.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.