

TCBS AGAR (VIBRIO SELECTIVE AGAR)
TM 436

For selective isolation of *Vibrio cholerae* & other enteropathogenic *Vibrio* causing food poisoning

Composition

Ingredients	g/L
Sucrose	20.00
Agar	15.00
Proteose peptone	10.00
Sodium citrate	10.00
Sodium thiosulphate	10.00
Sodium chloride	10.00
Ox bile	8.00
Yeast extract	5.00
Ferric citrate	1.00
Bromothymol blue	0.04
Thymol blue	0.04

*Dehydrated powder, hygroscopic in nature, store in a dry place in tightly - sealed containers below 25°C and protect from direct Sunlight.

Instructions for Use

Dissolve 89.0g in 1000ml distilled water. Gently heat to boil with gentle swirling and dissolve the medium completely. DO NOT AUTOCLAVE. Cool to 45 - 50°C and pour into sterile Petri plates.

Appearance: Bluish – green colour, clear to slightly opalescent gel

pH (at 25°C): 8.6 ± 0.2

Principle

TCBS AGAR (VIBRIO SELECTIVE AGAR) is highly selective for the isolation of *Vibrio cholerae* and *V. parahaemolyticus* from stool specimens, and specified in standard methods for food testing.

This highly selective medium meets the nutritional requirements of *Vibrio* species and allows competing with intestinal flora. *Vibrio* species is able to grow in media containing increased salt concentrations. *Vibrio* species are natural inhabitants of water.

Proteose peptone and Yeast extract are the sole sources of carbon, nitrogen, vitamin B - complex, minerals and amino acids in the medium. Sodium thiosulphate serves as a sulphur source and, in combination with Ferric citrate, detects hydrogen sulphide production. Sucrose is included as a fermentable carbohydrate for the metabolism of *Vibrio*. Inhibition of gram- positive bacteria is achieved by the incorporation of Ox bile, which is a synthetic occurring substance and suppresses primarily Enterococci. Ox bile also helps to inhibit gram - positive bacteria.

The alkaline pH of the medium enhances the recovery of *V. cholerae* because this organism is sensitive to acid environments. Thymol blue and Bromothymol blue are included as indicators of pH changes. Sodium citrate and Sodium thiosulphate are the selective agents, providing an alkaline pH to inhibit gram- positive organism and suppress coliforms. Agar is the solidifying agent. TCBS Agar deeps (pour



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tubes) are provided in a 20 ml fill so that the medium may be liquefied and poured into a petri plates. This provides a convenient source of medium with a longer shelf - life than pre- poured plated media.

Microbiological parameters (Growth promotion test)

Cultural characteristics observed after inoculation (10^3 CFU/ml) and incubation at 35 - 37°C for 18 - 48 hours.

Test strains	ATCC	Inoculum (CFU/ml)	Growth	Colony appearance
<i>Vibrio cholerae</i>	9459	10^3	Luxuriant	Yellow
<i>Vibrio parahemolyticus</i>	17802	10^3	Luxuriant	Bluish - green
<i>Vibrio alginolyticus</i>	17749	10^3	Luxuriant	Yellow

References

1. Department of Health NHS Executive: The Caldicott Committee. Report on the review of patient identifiable information. London. December. (1997).
2. Kobayashi, Enomoto, Sakazaki and Kuwahara, A new selective isolation medium for pathogenic vibrios: TCB5-Agar, Jap. J. Bacteriol., 18, 387. (1963).
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4. Kobayashi, T., S. Enomoto, R. Sakazaki, and S. Kuwahara. 1963. A new selective medium for pathogenic vibrios, TCBS (modified Nakanishi's agar). Jpn, J. Bacteriol. 18:387. Nakanishi, Y. (1963).