



TCBS AGAR

INTENDED USE

Thiosulfate Citrate Bile Salts Sucrose Agar (TCBS Agar) is used for the selective isolation of cholera vibrios and *Vibrio parahaemolyticus* from a variety of clinical and nonclinical specimens.^{1,2}

SUMMARY AND EXPLANATION

Vibrio species are most widely recognized for their role in human intestinal infections. Diarrheas caused by *Vibrio cholerae* and *V. parahaemolyticus* are important worldwide.³ The isolation of *Vibrio* species has been enhanced by the development of media which are highly selective for vibrios.

TCBS is the primary plating medium universally used for the selective isolation of vibrios that cause cholera, diarrhea and food poisoning. It was developed by Kobayashi et al.⁴, who modified the selective medium of Nakanishi.⁵ The combination of alkaline peptone water and TCBS Agar is used in many procedures for the isolation of *V. cholerae* and other *Vibrio* species from feces.¹⁻³

TCBS Agar Deeps (pour tubes) are provided in a 20 mL fill so that the medium may be liquefied and poured into a Petri dish. This provides a convenient source of medium with a longer shelf-life than pre-poured plated media.

PRINCIPLE

TCBS Agar is highly selective for the isolation of *V. cholerae* and *V. parahaemolyticus* as well as other vibrios. Inhibition of gram-positive bacteria is achieved by the incorporation of oxgall, which is a naturally occurring substance containing a mixture of bile salts, and sodium cholate, a pure bile salt. Sodium thiosulfate serves as a sulfur source and, in combination with ferric citrate, detects hydrogen sulfide production. Saccharose (sucrose) is included as a fermentable carbohydrate for the metabolism of vibrios. The alkaline pH of the medium enhances the recovery of *V. cholerae*. Thymol blue and bromthymol blue are included as indicators of pH changes.

REAGENTS (FORMULA)

Yeast Extract	5.0	g
Proteose Peptone No. 3	10.0	g
Sodium Citrate	10.0	g
Sodium Thiosulfate	10.0	g
Oxgall	8.0	g

Saccharose	20.0	g
Sodium Chloride	10.0	g
Ferric Ammonium Citrate	1.0	g
Bromthymol Blue	0.04	g
Thymol Blue	0.04	g
Agar	15.0	g
Deionized Water	1000.0	ml

PROCEDURE

Use standard procedures to obtain isolated colonies from specimens. Incubate the plates, protected from light, in an inverted position (agar side up) at 35°C for 24-48 hours.

EXPECTED RESULTS

Typical colonial morphology on TCBS Agar is as follows:

V. cholerae: Large yellow colonies.

V. parahaemolyticus: Colonies with blue to green centers.

V. alginolyticus: Large yellow colonies.

Proteus/Enterococci: Partial inhibition. If growth, colonies are small and yellow to translucent.

Pseudomonas/Aeromonas: Partial inhibition. If growth, colonies are blue.

QUALITY CONTROL

All lot numbers have been tested and have been found to be acceptable. Customers can test products using the following quality control organisms. Testing of control organisms should be performed in accordance with established laboratory quality control procedures. If aberrant quality control results are noted, sample results should not be reported.

Organisms	Incubation	Results
<i>Escherichia coli</i> ATCC 25922	35 ± 2°C for 18-24 hours	Not Growth
<i>Vibrio alginolyticus</i> ATCC 17749	35 ± 2°C for 18-24 hours	Growth, Yellow Colony
<i>Vibrio parahemolyticus</i> ATCC 17802	35 ± 2°C for 18-24 hours	Growth, Blue green Colony

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BIBLIOGRAPHY

- Downes and Ito. 2001. Compendium of methods for the microbiological examination of foods, 4th ed. American Public Health Association, Washington, D.C.
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3. Murray, Baron, Jorgensen, Landry and Pfaller (ed.). 2007. Manual of clinical microbiology, 9th ed. American Society for Microbiology, Washington, D.C.
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5. Nakanishi. 1963. Modern Media 9: 246.



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