

# T.C.B.S. Cholera Medium

### (Thiosulphate Citrate Bile Salts Sucrose Agar)

## LAB 96

#### Description

T.C.B.S. is designed for the selective isolation of *Vibrio* species, particularly *V. cholerae*. The formulation was developed by Kobayashi, Enomoto, Sakazaki and Kuwahara and inhibits most of the *Enterobacteriaceae* for at least 24 hours. Therefore heavy inoculation of the medium is possible.

Formula	g/litre
Yeast Extract	5.5
Peptone Mix	10.0
Sodium thiosulphate	10.0
Sodium citrate	10.0
Bile salts	9.0
Sucrose	17.0
Sodium chloride	10.0
Ferric citrate	1.0
Bromothymol blue	0.04
Thymol blue	0.04
Agar No. 1	15.0

#### Method of reconstitution

Weigh 88 grams of powder and add to 1 litre of deionised water. Allow to soak for 10 minutes, swirl to mix then bring to the boil. Cool to 47°C and pour into Petri dishes. DO NOT AUTOCLAVE OR OVERHEAT THIS MEDIUM.

Appearance: Dark green clear agar.

**pH:** 8.6 ± 0.2

<u>Minimum Q.C. organisms:</u> V. cholerae (type F) E. coli (inhibition) NCIMB 50034

Storage of Prepared Medium: Plates - up to 7 days at 4°C in the dark. Capped containers - up to 1 month at 15-20°C in the dark.

Inoculation: Surface plating with a heavy inoculum, streak out to single colonies.

Incubation: 37°C aerobically for 18-24 hours.

Growth Characteristics					
organism	colony size (mm)	shape & surface	colour	other	
Vibrio. cholerae	2.0-3.0	CV.E.G.	Yellow	may revert to green at R.T.	
V. parahaemolyticus	3.0-5.0	CV.E.G.	Blue or Green		
V. alginolyticus	3.0-5.0	CV.E.G.	Yellow		
V. metschnikovii	2.0-4.0	CV.E.G.	Yellow		
V. fluvialis	2.0-3.0	CV.E.G.	Yellow		
V. vulnificus	2.0-3.0	CV.E.G.	Yellow		
V. mimicus	2.0-3.0	CV.E.G.	Green		
Enterococci	1.0	CV.E.G.	Yellow		
Proteus spp.	1.0	F.CR.G.	Green/Yellow		
Ples. shigelloides	P.P.		Green		

#### References

Kobayashi, T., Enomoto, S., Sakazaki, R. and Kuwahara, S. (1963). Jap. Bacteriol 18: 10-11, 387-391.

Furniss, A.L., Lee J.V. and Donovan, T.J. (1978). The Vibrios, PHLS Monograph No. 11.