Triple Sugar Iron Agar

LAB 53

Description

This is a modification of the Krumwiede and Kohn medium of 1917 which differentiates some of the *Enterobacteriaceae* on the basis of four reactions; fermentation of lactose, glucose and sucrose and H_2S production. This medium should be used in conjunction with a urease test to eliminate *Proteus* spp. when screening for *Salmonella* spp.

Formula	g/litre
Beef Extract	3.0
Yeast Extract	3.0
Balanced Peptone No. 1	20.0
Sodium chloride	5.0
Lactose	10.0
Sucrose	10.0
Glucose	1.0
Ferric citrate	0.3
Sodium thiosulphate	0.3
Phenol red	0.025
Agar No. 2	12.0

Method for reconstitution

Weigh 65 grams of powder, disperse in 1 litre of deionised water. Allow to soak for 10 minutes, swirl to mix then bring to the boil with frequent swirling to dissolve the solids. Distribute into tubes and sterilise at 121°C for 15 minutes. Allow to set as a slope ensuring that the slant is over a butt approximately 3cm deep.

Appearance: Reddish-orange gel.

pH: 7.4 ± 0.2

Minimum Q.C. organisms: *E. coli* NCIMB 50034
Ps. aeruginosa NCIMB 50067

Storage of Prepared Medium: Capped containers – up to 3 months at 15-20°C in the dark.

Inoculation: A heavy inoculum is streaked over the surface of the slope and stabbed into the butt.

Incubation: 37°C aerobically for 24 hours.

Interpretation		
Slant/butt	**Colour**	**Utilisation**
Alkaline/acid	Red/yellow	Glucose only fermented Peptones utilised
Acid/acid	Yellow/yellow	Glucose fermented Lactose + or sucrose fermented
Alkaline/alkaline	Red/Red	Neither glucose, lactose, nor sucrose fermented Peptones utilised

Organism	Butt	Slant	Sulphide production
Shigella dysenteriae *S. sonnei* *S. flexneri*	Acid or Alk.	NC or Alk.	-
Salmonella typhi	Acid	NC	+
S. paratyphi *S. cholerasuis*	Acid Gas	NC	-
S. typhimurium *S. enteritidis* *S. pullorum*	Acid Gas	NC	+
S. gallinarum	Acid	NC	+
E. coli *Enterobacter aerogenes* *E. cloacae*	Acid Gas	Acid	-
Proteus mirabilis	Acid Gas	Acid	+
Providencia rettgeri	Acid	NC	-

NC = No Change

References

American Public Health Association (1963). Diagnostic Procedures and Reagents, 4th edn., A.P.H.A., New York.

American Public Health Association (1966). Recommended Methods for the Microbiological Examination of Foods. 2nd edn., A.P.H.A., New York.

Edwards, P.R. and Ewing, W.H. (1962). Identification of *Enterobacteriaceae.* Burgess Publishing Co., Minneapolis.