

14703 *Cronobacter* spp. **ChromoSelect Agar, Modified** (Modified; *Cronobacter* spp. isolation agar; CSIA; *Enterobacter sakazakii* isolation agar; ESIA)

Cronobacter spp. **ChromoSelect Agar** modified meets the formulation recommended by ISO Committee for the isolation and identification of *Cronobacter* spp. (formerly *Enterobacter sakazakii*) from milk and milk products.

Composition:

Ingredients	Grams/Litre
Casein enzymic hydrolysate	7.0
Yeast extract	3.0
Sodium chloride	5.0
Sodium deoxycholate	0.6
Chromogenic substrate	0.15
Crystal violet	0.002
Agar	15.0
Final pH 7.0 +/- 0.2 at 25°C	

Store prepared media below 8°C, protected from direct light. Store dehydrated powder, in a dry place, in tightly-sealed containers at 2-8°C.

Appearance: Pinkish beige coloured, homogeneous, free flowing powder.

Gelling: Firm

Color and Clarity: Purple coloured, clear to slightly opalescent gel forms in petri plates.

Directions:

Suspend 30.75 g in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C and pour into sterile petri plates.

Principle and Interpretation:

Cronobacter species are ubiquitous and frequently found on vegetable, meat, fermented bread, dairy products and especially in baby food. *Cronobacter* spp. has been closely associated with neonatal meningitis and sepsis (1). This medium is recommended by ISO Committee for the isolation and identification of *Cronobacter* spp. (2). The chromogenic substrate is cleaved specifically (3) by *Cronobacter* spp. resulting in blue green colonies. Other organisms, which do not cleave this substrate produce colourless to slightly violet coloured colonies. Incorporation of the chromogenic mixture in the media renders an intense blue to blue-green colour to *Cronobacter* species colonies whereas *Enterobacter* species give colourless or blue centered colonies.

Casein enzymic hydrolysate and Yeast extract provide the essential growth nutrients like vitamins, minerals and B-complex vitamin along with nitrogenous and carbonaceous compounds. Sodium chloride helps in maintaining the osmotic equilibrium of the medium and sodium deoxycholate inhibits the accompanying gram positive flora. Crystal violet inhibits as well gram-positive microorganisms especially Staphylococci.



Cultural characteristics after 18-24 hours at 44±1°C.

Organisms (ATCC)	Growth	Color of Colony
<i>Escherichia coli</i> (25922)	+++	colourless
<i>Enterobacter aerogenes</i> (13048)	+++	colourless with blue centre
<i>Enterobacter faecalis</i> (29212)	-	-
<i>Cronobacter sakazakii</i> (12868)	+++	blue-green
<i>Staphylococcus aureus</i> (25923)	-	-

References:

1. H.L. Muytjens, H.C. Zanen, H.J. Sonderkamp et al., Analysis of eight cases of neonatal meningitis and sepsis due to *Enterobacter sakazakii*, J. Clin. Microbiol. 18,115-120 (1983)
2. International Organization for Standardization Draft ISO/ TS 22964, 2006 (E)
3. Isenberg (ed.), Clinical microbiology procedures handbook, vol. 1. American Society for Microbiology, Washington, DC (1992)

Precautions and Disclaimer

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