B.C.P. - D.C.L.S. Agar

M219

B.C.P.-D.C.L.S. Agar (Bromo Cresol Purple- Deoxycholate -Citrate-Lactose- Sucrose Agar) is used for the selective isolation of *Salmonella* and *Shigella* species

Composition**

Ingredients	Gms / Litre
Peptic digest of animal tissue	5.000
Casein enzymic hydrolysate	5.000
Yeast extract	3.000
Beef extract	3.000
Lactose	7.500
Sucrose	7.500
Sodium citrate	10.000
Sodium chloride	5.000
Sodium thiosulphate	5.000
Sodium deoxycholate	2.500
Bromocresol purple	0.020
Agar	14.000
Final pH (at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 67.52 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. DO NOT AUTOCLAVE or OVERHEAT. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

Principle And Interpretation

Salmonella and *Shigella* are gram-negative, facultatively anaerobic, non-sporulating rods in the family *Enterobacteriaceae* . They are widely distributed in animals affecting mainly the stomach and the intestines. *Shigella* is the causative agent of bacterial diarrhoea and the faecal-oral route usually transmits the disease. Human *Salmonella* infections are most commonly caused by ingestion of food, water or milk contaminated by human or animal excreta (1). Arizona group was originally named *Salmonella* Arizonae. It has been found mainly in reptiles and birds and occasionally in human patients with diarrhoea or septicemia. These organisms are difficult to differentiate biochemically from *Escherichia coli* , one of the most commonly recovered bacteria in clinical laboratory.

B.C.P-D.C.L.S Agar (Bromo Cresol Purple - Deoxycholate - Citrate - Lactose-Sucrose Agar) is the modification of the original formulation of Leifson (2), which was later, modified by Hajna and Damon (3). It allows easy isolation of *Salmonella* , *Shigella* and *Arizona* organisms from a mixed culture by differentiating between lactose-negative, sucrose-positive coliforms. It also inhibits all gram-positive bacteria and most of the *Proteus* species along with some strains of *S. dysentriae* (4).

Larger amount of the material can be inoculated into an enrichment medium followed by inoculation onto an agar plate, thereby, facilitating the isolation of Salmonella, when present only in small numbers. On incubation, *Salmonella* multiply rapidly, while *E.coli* and most other bacteria are inhibited. After enrichment, the enriched culture is plated onto a differential agar medium. B.C.P.-D.C.L.S. is a useful modification of D.C.A. (Deoxycholate Citrate Agar) that contains both lactose and sucrose (3). Some coliforms ferment sucrose more readily than lactose. Sucrose fermenting and lactose non-fermenting strains, e.g. some strains of *Proteus* and *E.coli* , form colonies distinguishable from the pale colonies of *Salmonella* and *Shigella* , which do not ferment sucrose, on this medium. Hence the number of false positive cultures requiring biochemical testing is reduced and the efficiency of isolation of *Salmonella* and *Shigella* is increased.

Peptic digest of animal tissue, casein enzymic hydrolysate, yeast extract and beef extract in the medium provide nitrogen, vitamins and minerals necessary to support bacterial growth. Lactose and sucrose are the fermentable carbohydrates and therefore inclusion of these two sugars permits the formation of yellow colonies by organisms that ferment lactose, sucrose or both. Sodium thiosulphate is the indicator of H2S production. Sodium citrate and sodium deoxycholate inhibit all gram-positive bacteria and coliforms but allow the gram-negative bacilli to grow. Sodium chloride provides essential ions. Bromo cresol purple is the pH indicator.

B.C.P.-D.C.L.S. Medium is unsuitable for the isolation of *Yersinia* species, which are sucrose positive. Non-selective media should be inoculated along with this media.

The medium can be directly inoculated with the test specimens. Alternatively, the sample can be enriched in GN Broth, Hajna (M242), Tetrathionate Broth (M032), or Selenite Broth (M052), and subsequently isolated on B.C.P.-D.C.L.S. Agar. A less inhibitory medium should be run in parallel to B.C.P.-D.C.L.S.

Quality Control

Appearance
Light yellow to beige homogeneous free flowing powder

Gelling
Firm,comparable with 1.4% Agar gel.

Colour and Clarity of prepared medium
Purple coloured, clear to slightly opalescent gel forms in Petri plates.

Reaction
Reaction of 6.75% w/v aqueous solution at 25°C. pH : 7.2±0.2

pH
7.00-7.40

Cultural Response
M219: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony
Cultural Response				
Escherichia coli ATCC 25922	50-100	none-poor	<=10%	yellow
Proteus mirabilis ATCC 25933	50-100	none-poor	<=10%	colourless
Proteus vulgaris ATCC 13315	50-100	none-poor	<=10%	colourless
Salmonella Typhimurium ATCC 14028	50-100	good-luxuriant	>=50%	colourless, may show faint bluish coloured colonies
Salmonella Enteritidis ATCC 13076	50-100	good-luxuriant	>=50%	colourless, may show faint bluish coloured colonies
Shigella dysenteriae ATCC 13313	50-100	good	>=50%	colourless, may show faint bluish coloured colonies
Shigella flexneri ATCC 12022	50-100	good-luxuriant	>=50%	colourless, may show faint bluish coloured colonies
Shigella sonnei ATCC 25931	50-100	good-luxuriant	>=50%	colourless, may show faint bluish coloured colonies

Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label.

Reference

1. Koneman E. W., Allen S. D., Janda W. M., Schreckenberger P. C., Winn W. C. Jr., 1992, Colour Atlas and Textbook of Diagnostic Microbiology, 4th Ed., J. B. Lippincott Company.
2. Leifson E., 1935, J. Pathol. Bacteriol. 40, 581
3. Hajna A. A. and Damon S. R., 1956, Appl. Microbiol., 4, 341
4. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. I, Williams and Wilkins, Baltimore.

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