HiCombi™ Dual Performance Salmonella Medium - DCA

LQ031A

Intended use

Recommended as a qualitative test for rapid growth and confirmation of *Salmonella*. Combination of solid (20ml) and liquid (40 ml) media in single bottle.

Composition**

Ingredients	g / L
Solid	20.000ml
HI solids #	10.000
Proteose peptone	10.000
Lactose	10.000
Sodium deoxycholate	5.000
Neutral red	0.020
Sodium citrate	20.000
Ferric ammonium citrate	2.000
Agar	13.500
Liquid	40.000 ml

Same as solid media without Agar
**Formula adjusted, standardized to suit performance parameters
Equivalent to Heart Infusion solids

Directions

Label the ready to use LQ031A bottle. Remove the top seal of the cap. Disinfect the part of the rubber stopper which is now exposed. Transfer the sample immediately into the culture bottle by puncturing the rubber stopper with the needle. Venting: Use sterile venting needle (LA038). Keep the bottle in an upright position preferably in a biological safety cabinet, place an alcohol swab over the rubber stopper and insert the venting needle with filter through it. Insertion and withdrawal of the needle should be done in a straight line. Discard the needle and mix the contents by gently inverting the bottle 2-3 times. Do not vent the bottle for anaerobic cultures. Incubate at 35-37°C for 18-24 hours. Recommended volume of blood to be tested in LQ031A: 8-10 ml (For Adult use).

Principle And Interpretation

Deoxycholate Citrate Agar is prepared as per the modified formula of Leifson (1). This medium is used for the isolation and maximum recovery of intestinal pathogens belonging to *Salmonella* and *Shigella* groups from foods (2). However, it is recommended to use less inhibitory medium when *Shigella* have to be isolated (3). The selectivity of this medium permits the use of fairly heavy inocula without danger of overgrowth of *Shigella* and *Salmonella* by other microflora. This medium is similar to deoxycholate agar in comparison but is moderately more selective for enteric pathogens owing to increased concentrations of both citrate and deoxycholate salts. Sodium deoxycholate at pH 7.3 to 7.5 is inhibitory for gram-positive bacteria. Citrate salts, in the concentration included in the formulation, are inhibitory to gram-positive bacteria and most other normal intestinal organisms.

HI solids is a source of carbon and nitrogen and this ingredient is used because the inhibition of coliforms produced is greater than when an extract or simple peptone is used. Proteose peptone provides carbon, nitrogen, vitamins and minerals. Coliform bacteria and gram-positive bacteria are inhibited or greatly suppressed due to sodium deoxycholate, sodium citrate and ferric ammonium citrate. Lactose helps in differentiating enteric bacilli, as lactose fermenters produce red colonies while lactose non-fermenters produce colourless colonies. Coliform bacteria, if present form pink colonies on this medium. The degradation of lactose causes acidification of the medium surrounding the relevant colonies and the pH indicator neutral red changes its colour to red. These colonies usually are also surrounded by a turbid zone of precipitated deoxycholic acid due to acidification of the medium. Sodium deoxycholate combines with neutral red in an acidic environment, causing the dye to go out of the solution with the subsequent precipitation of deoxycholate (1).

The reduction of ferric ammonium citrate to iron sulfide is indicated by the formation of black iron sulfide. *Salmonella* and *Shigella* species do not ferment lactose but *Salmonella* may produce H_2S, forming colorless colonies with or without black

centers. Citrate and iron (Fe) combination has a strong hydrolyzing effect on agar when the medium is heated, producing a soft and unelastic agar. If autoclaved the agar becomes soft and almost impossible to streak. *Salmonella* gallinarum is inhibited if sodium deoxycholate concentration is increased to 0.1% or greater (1). Surface colonies of non-lactose fermenters often absorb a little colour (pinkish) from the medium and organisms may be mistaken for coliforms (1).

Type of specimen

Clinical- Blood

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (4,5).
After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

In Vitro diagnostic use only. For professional use only. Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling clinical specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Further biochemical identification is required for confirmation of species.
2. Individual organisms differ in their growth requirement and may show variable growth patterns on the medium.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance
In a sterile glass bottle combination of broth and one agar coated surface.

Colour of Agar medium
Reddish orange coloured medium

Colour of liquid medium
Red coloured medium

Quantity of medium
20ml of solid medium in glass bottle 40ml of liquid medium in glass bottle

pH of Agar medium
7.30-7.70

pH of liquid medium
7.20-7.60

Sterility Check
Passes release criteria

Cultural response
Cultural characteristics was observed after incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth on agar medium	Growth on liquid medium	Colour of colony
Enterococcus faecalis ATCC 29212 (00087*)	>= 10^4	Inhibited	Inhibited	-
Escherichia coli ATCC 25922 (00013*)	50-100	Poor	Luxuriant	Pink w/ bile precipitate
Salmonella Enteritidis ATCC 13076 (00030*)	50-100	Luxuriant	Luxuriant	Colourless colonies with black center(H_2S production)
Salmonella Typhimurium ATCC 14028 (00031*)	50-100	Luxuriant	Luxuriant	Colourless colonies with black center(H_2S production)
Shigella flexneri ATCC 12022 (00126*)	50-100	Good	Luxuriant	Colourless
Escherichia coli ATCC 8739 (00012*)	50-100	Poor	Luxuriant	Pink w/ bile precipitate
Salmonella Abony NCTC 6017 (00029*)	50-100	Luxuriant	Luxuriant	Colourless colonies with black center(H_2S production)
Staphylococcus aureus subsp. aureus ATCC 25923 (00034*)	>=10^4	Inhibited	Inhibited	

Key : (*) Corresponding WDCM numbers.

Storage and Shelf Life

On receipt store between 2-8°C. Use before expiry date on the label. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (4,5).

Reference

1. Leifson, 1935, J. Path. Bact., 40:581.
2. Salfinger Y., and Tortorello M.L., 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
3. Frieker C.R., 1987, J. Appl. Bact., 63:99.
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
5. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

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HiMedia Laboratories Pvt. Limited,
Plot No.C-40, Road No.21Y,
MIDC, Wagle Industrial Area,
Thane (W) -400604, MS, India

IVD ***In vitro* diagnostic medical device**

2°C 8°C **Storage temperature**

EC REP CEpartner4U, Esdoornlaan 13,
3951DB Maarn, NL
www.cepartner4u.eu

CE Marking

Do not use if package is damaged