Deoxycholate Agar

M030

Intended Use:

Deoxycholate Agar is used as a differential medium for the direct count of coliforms in dairy products. Also used for the isolation of enteric pathogens from rectal swabs, faeces and other pathological specimens.

Composition**

Ingredients	g / L
Peptone	10.000
Lactose	10.000
Sodium deoxycholate	1.000
Sodium chloride	5.000
Dipotassium hydrogen phosphate	2.000
Ferric citrate	1.000
Sodium citrate	1.000
Neutral red	0.030
Agar	15.000
Final pH (at 25°C)	7.3±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 45.03 grams in 1000 ml purified/distilled water. Heat to boiling to dissolve the medium completely. DO NOT AUTOCLAVE. Avoid excessive or prolonged heating during reconstitution. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

Principle And Interpretation

Deoxycholate Agar is prepared as per the formulation by Leifson (1). This media is used for the isolation and maximum recovery of intestinal pathogens belonging to *Salmonella* and *Shigella* species (2). The selectivity of medium permits the use of fairly heavy inocula without danger of overgrowth of the *Shigella* and *Salmonella* by other micro-flora. For the routine examination of stool and urine specimens, it is recommended that other media such as MacConkey Agar (M082), Bismuth Sulphite Agar (M027) etc. be used in conjunction with this medium. It can also be used to streak specimen from Selenite Broth cultures. This is particularly recommended for the detection of *Shigella* and *Salmonella* in the examination of rectal swabs and faeces. These organisms produce colourless colonies on this medium.

Peptone provides carbon, nitrogen, long chain amino acids, vitamins and minerals. Coliform bacteria and gram-positive bacteria are inhibited or greatly suppressed due to sodium deoxycholate and sodium citrate. Sodium chloride maintains the osmotic balance of the medium while dipotassium phosphate buffers the medium. 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 (3).

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. Surface colonies of non-lactose fermenters often absorb a little colour (pinkish) from the medium and organisms may be mistaken for coliforms (2).

Type of specimen

Clinical : Stool and urine specimens, etc; Food and Dairy samples

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines(4,5). For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (6). After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

In Vitro diagnostic use. 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.The medium is highly selective and hence is recommended to be used in conjunction with less selective medium like MacConkey Agar.
2.The medium is not recommended, when low recovery of the desired pathogen is expected.

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
Light yellow to pink homogeneous free flowing powder
Gelling
Firm, comparable with 1.5% Agar gel
Colour and Clarity of prepared medium
Reddish orange coloured, clear to slightly opalescent gel forms in Petri plates
Reaction
Reaction of 4.5% w/v aqueous solution at 25°C. pH : 7.3±0.2
pH
7.10-7.50
Cultural Response
Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Colour of Colony
Salmonella Typhi ATCC 6539	50-100	luxuriant	>=50%	colourless
Staphylococcus aureus subsp. *aureus* ATCC 25923 (00034*)	>=10^4	inhibited	0%	
Enterococcus faecalis ATCC 29212 (00087*)	>=10^4	inhibited	0%	
Escherichia coli ATCC 25922 (00013*)	50-100	good	40-50%	pink with bile precipitate
Salmonella Enteritidis ATCC 13076 (00030*)	50-100	good-luxuriant	>=50%	colourless
Salmonella Typhimurium ATCC 14028 (00031*)	50-100	good-luxuriant	>=50%	colourless
Shigella flexneri ATCC 12022 (00126*)	50-100	good	40-50%	colourless

Key : (*) Corresponding WDCM numbers.

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. 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. Lapage S., Shelton J. and Mitchell T., 1970, Methods in Microbiology', Norris J. and Ribbons D., (Eds.), Vol. 3A, Academic Press, London.
2. MacFaddin J. F., 2000, Biochemical Tests for Identification of Medical Bacteria, 3rd Ed., Lippincott, Williams and Wilkins, Baltimore.
3. Leifson, 1935, J. Path. Bacteriol., 40:581.
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.
6. 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.

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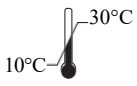
IVD ***In vitro* diagnostic medical device**



Storage temperature

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

CE Marking

Do not use if package is damaged

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