

LEIFSON Agar

(Deoxycholate Citrate Agar acc. to LEIFSON, modified)

Medium proposed by LEIFSON (1935) and modified by HYNES (1942) for the isolation of salmonellae and shigellae.

The culture medium complies with the European Pharmacopeial.

Mode of Action

The concentrations of deoxycholate and citrate contained in this medium are so high that they completely suppress the gram-positive microbial flora and inhibit the coliform bacteria more or less. Salmonellae grow normally; some species of *Shigella* are slightly inhibited (e.g. *Shig. shigae*).

The degradation of lactose causes an 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. Colonies of lactose-negative microorganisms are colourless. The reduction of thiosulfate to sulfide is indicated by the formation of black iron sulfide.

Typical Composition (g/litre)

Meat extract 5.0; peptone from meat 5.0; lactose 10.0; sodium thiosulfate 5.4; ammonium iron(III) citrate 1.0; sodium citrate 6.0; sodium deoxycholate 3.0; neutral red 0.02; agar-agar 12.0:

Preparation

Suspend 47.5 g/litre, cool quickly, pour plates.

The medium is heat sensitive. Bring to boil with frequent agitation. Do not remelt.

■ Do not autoclave.

pH: 7.5 ± 0.2 at 25 °C.

The plates are clear and reddish-brown.

■ The prepared culture medium can be stored for 1 week in the refrigerator.

Experimental Procedure and Evaluation

Inoculate by spreading the sample or material from an enrichment culture on the surface of the culture medium.

In view of the strong inhibitory action of LEIFSON Agar, it is advisable to use a less inhibitory selective medium as well, e.g. MaxCONKEY Agar or Deoxycholate Lactose Agar.

Incubation: 24-48 hours at 35 °C aerobically.

| Appearance of Colonies | Microorganisms |
|--|---|
| After 18 hours: pale pink to colourless, diameter 1 mm. After 48 hours: slightly opaque, often with a central gray dot, diameter approx. 2 mm | <i>Salmonella typhosa</i> |
| After 18 hours: pale pink to colourless, diameter approx. 1mm After 48 hours: slightly opaque, convex, with a central black dot | <i>Salmonella paratyphi B</i> and other H ₂ S-positive salmonellae |
| At first colourless, then pale pink (slight lactose degradation). After 18 hours: flat, diameter approx. 1 mm. After 38 hours: diameter approx 2 mm | <i>Shigella sonnei</i> |
| As <i>S. sonnei</i> but with a convex centre, often with flat edges | <i>Shigella flexneri</i> |
| Similar to <i>Salmonella</i> and <i>Shigella</i> , characteristic sweet smell | <i>Pseudomonas</i> |
| Similar to <i>Salmonella</i> and <i>Shigella</i> , black central dot | <i>Proteus vulgaris</i> , most strains of <i>Proteus mirabilis</i> |
| Inhibited growth, pink colonies surrounded by turbid precipitation zone, diameter approx. 1-2 mm | <i>Escherichia coli</i> |
| Inhibited growth, colourless or pink centre, convex, mucoid, opaque, diameter approx. 1-2mm | <i>Enterobacter</i> , <i>Klebsiella</i> |

Literature

European Pharmacopeia II, Chapter VIII, 10.

LEIFSON, E.: New culture media based on sodium deoxycholate for the isolation of intestinal pathogens and for the enumeration of colon bacilli in milk and water. - *J. Path. Bact.*, **40**; 581-599 (1935).

HYNES, M.: The isolation of intestinal pathogens by selective media. -*J.Path. Bact.*, **54**; 193-207 (1942).

Ordering Information

| Product | Merck Cat. No. | Pack size |
|---|----------------|-----------|
| LEIFSON Agar (Deoxycholate Citrate Agar acc. to LEIFSON, modified) | 1.02896.0500 | 500 g |
| MacCONKEY Agar | 1.05465.0500 | 500 g |
| XLD Agar | 1.05287.0500 | 500 g |

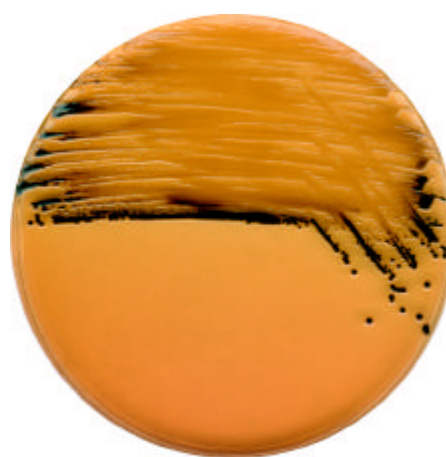
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Quality control

| Test strains | Growth | Colour | Colonies Precipitate | Black centre |
|--|------------------|------------|----------------------|--------------|
| <i>Escherichia coli</i> ATCC 25922 | poor / fair | red / pink | + | - |
| <i>Klebsiella pneumoniae</i> ATCC 13883 | good / very good | red / pink | - | - |
| <i>Shigella flexneri</i> ATCC 12022 | fair / very good | colourless | - | - |
| <i>Shigella sonnei</i> ATCC 11060 | fair / very good | colourless | - | - |
| <i>Salmonella typhimurium</i> ATCC 14028 | good / very good | colourless | - | + |
| <i>Salmonella enteritidis</i> NCTC 5188 | good / very good | colourless | - | + |
| <i>Proteus mirabilis</i> ATCC 14273 | good / very good | colourless | - | ± |
| <i>Staphylococcus aureus</i> ATCC 25923 | none | | | |
| <i>Bacillus cereus</i> ATCC 11778 | none | | | |



Proteus mirabilis
ATCC 14273



Salmonella enteritidis
NCTC 5188