

**Lead Acetate Agar****M180**

Lead Acetate Agar is recommended for the detection of hydrogen sulphide producing enteric bacteria.

**Composition\*\*\***

Ingredients	Gms / Litre
Peptic digest of animal tissue	15.000
Proteose peptone	5.000
Dextrose	1.000
Lead acetate	0.200
Sodium thiosulphate	0.080
Agar	15.000
Final pH ( at 25°C)	6.6±0.2

\*\*Formula adjusted, standardized to suit performance parameters

**Directions**

Suspend 36.28 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Dispense into test tubes and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Allow the tubes to cool in a slanted position to obtain slants with generous butts. Inoculate pure culture by surface streaking the slant and stabbing the butt.

**Principle And Interpretation**

*Salmonella*, *Shigella*, *Yersinia* species and certain strains of *Escherichia coli* cause severe gastroenteritis and life-threatening systemic illness in human (1, 2). Of these, *Salmonella* Typhi can be differentiated due to their ability to form hydrogen sulphide (3). Lead Acetate Agar is the modification of the original formulation of Spray (4). This medium was successfully used to study hydrogen sulphide production (4, 5). Lead Acetate Agar can also be used to differentiate between *Salmonella* Paratyphi A and *Salmonella* Paratyphi B (6). The latter produces hydrogen sulphide, observed as browning of the medium, within 18-24 hours, whereas the former fails to produce hydrogen sulphide.

Peptic digest of animal tissue, proteose peptone and dextrose provide all the essential nutrients for the growth of bacteria. Bacteria capable of using sulphur from sodium thiosulphate in their metabolic activities produce hydrogen sulphide. Lead acetate acts as an indicator of hydrogen sulphide production observed as browning of the medium. Dextrose is the fermentable carbohydrate source. Production of gas from dextrose is indicated by the presence of bubbles in the butt.

**Quality Control****Appearance**

Cream to yellow homogeneous free flowing powder

**Gelling**

Firm, comparable with 1.5% Agar gel

**Colour and Clarity of prepared medium**

Medium amber coloured clear to slightly opalescent gel forms in tubes as slants

**Reaction**

Reaction of 3.63% w/v aqueous solution at 25°C. pH : 6.6±0.2

**Cultural Response**

M180: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Gas Production	H <sub>2</sub> S production		
<i>Escherichia coli</i> ATCC 25922	50-100	luxuriant	positive reaction	negative reaction		
<i>Enterobacter aerogenes</i> ATCC 13048	50-100	luxuriant	positive reaction	negative reaction		
<i>Salmonella</i> Paratyphi A ATCC 9150	50-100	luxuriant	negative reaction	negative reaction		
<i>Salmonella</i> Paratyphi B ATCC 8759	50-100	luxuriant	negative reaction	positive reaction, browning of the medium		
<i>Salmonella</i> Typhi ATCC 6539	50-100	luxuriant	variable reaction	positive reaction,		

				browning of the medium		
<i>Salmonella Typhimurium</i> ATCC 14028	50-100	luxuriant	negative reaction	positive reaction, browning of the medium		
<i>Shigella dysenteriae</i> ATCC 13313	50-100	luxuriant	negative reaction	negative reaction		
<i>Shigella flexneri</i> ATCC 12022	50-100	luxuriant	negative reaction	negative reaction		

**Reference**

1. Balows A., Hausler W. J. Jr., Hermann K. L., Isenberg H. D., Shadomy H. J., (Eds.), Manual of Clinical Microbiology, 5th Ed., ASM, Washington, D.C.
2. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Tenover F. C., (Ed.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.
3. Orłowski, 1897, Dissert, St. Petersburg.
4. Spray R. S., 1936, J. Bacteriol., 32:135.
5. Morrison L. E. and Tanner F. W., 1922, J. Bacteriol., 7:343.
6. Jordan E. O. and Victorson R., 1917, J. Infect. Dis., 21:554.

**Storage and Shelf Life**

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