



## Endo Agar Base

M1077

Endo Agar Base is recommended for preparing Endo Agar to confirm presumptive test for lactose fermenting coliforms.

### Composition\*\*

Ingredients	Gms / Litre
Peptic digest of animal tissue	10.000
Lactose	10.000
Dipotassium phosphate	3.500
Sodium sulphite	2.500
Agar	12.000
Final pH ( at 25°C)	7.5±0.2

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

### Directions

Suspend 38 grams in 1000 ml distilled water. Add 4 ml of 10% Basic Fuchsin (FD059). Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Mix well before pouring into sterile Petri plates.

If the solidified culture medium is somewhat too red, then to remove the colour, add a few drops (max. 1 ml/litre) of a freshly prepared 10% Sodium sulphite solution and boil.

*Caution : Basic fuchsin is a potential carcinogen and care should be taken to avoid inhalation of the powdered dye and contamination of the skin .*

### Principle And Interpretation

Endo had first developed a culture medium for differentiation of lactose fermenters and lactose non-fermenters while inhibiting gram-positive bacteria (1). Inhibition of the later was achieved without the use of bile salts as was traditionally used. Endo was successful in inhibiting gram-positive bacteria on his medium by the incorporation of sodium sulphite and basic fuchsin. The resulting Endo Agar, also known as Fuchsin Sulphite and Infusion Agar, was used to isolate the typhoid bacilli. Many modifications of this media have been done over the years. Endo Agar is recommended by APHA as an important medium in the microbiological examination of water and wastewater, dairy products and foods (2-4). Endo Agar is used to confirm the detection and enumeration of coliform bacteria following presumptive test of drinking water. It is also used for the detection and isolation of coliforms and fecal coliforms from milk, dairy products and food.

The medium contains peptic digest of animal tissue which provide nitrogen, carbon, vitamins and minerals required for bacterial growth. Sodium sulphite and basic fuchsin (FD) has inhibitory effect on gram-positive microorganisms. Lactose fermenting coliforms produce aldehyde and acid. The aldehyde in turn liberates fuchsin from the fuchsin-sulphite complex, giving rise to a red colouration of colonies. With *Escherichia coli* , this reaction is very pronounced as the fuchsin crystallizes, exhibiting a permanent greenish metallic lustre (fuchsin lustre) to the colonies.

### Quality Control

#### Appearance

Cream to yellow homogeneous free flowing powder

#### Gelling

Firm, comparable with 1.2% Agar gel

#### Colour and Clarity of prepared medium

After addition of FD059: Orangish pink coloured, After addition of FD059: clear to slightly opalescent gel with fine precipitate forms in Petri plates.

#### Reaction

Reaction of 3.8% w/v aqueous solution at 25°C. pH : 7.5±0.2

#### pH

7.30-7.70

**Cultural Response**

Cultural characteristics observed with added Basic fuchsin (FD059) after an incubation at 35-37°C for 18-24 hours.

**Cultural Response**

Organism	Inoculum (CFU)	Growth	Recovery	Colour of Colony
<b>Cultural Response</b>				
<i>Bacillus subtilis</i> ATCC 6633	$\geq 10^3$	inhibited	0%	
<i>Enterobacter aerogenes</i> ATCC 13048	50-100	good-luxuriant	$\geq 50\%$	pink
<i>Enterococcus faecalis</i> ATCC 29212	50-100	none-poor	$\leq 10\%$	pink, small
<i>Escherichia coli</i> ATCC 25922	50-100	good-luxuriant	$\geq 50\%$	pink to rose red with metallic sheen
<i>Klebsiella pneumoniae</i> ATCC 13883	50-100	good-luxuriant	$\geq 50\%$	pink, mucoid
<i>Proteus vulgaris</i> ATCC 13315	50-100	good-luxuriant	$\geq 50\%$	colourless to pale pink
<i>Pseudomonas aeruginosa</i> ATCC 27853	50-100	good-luxuriant	$\geq 50\%$	colourless, irregular
<i>Salmonella Typhi</i> ATCC 6539	50-100	good-luxuriant	$\geq 50\%$	colourless to pale pink
<i>Shigella sonnei</i> ATCC 25931	50-100	good-luxuriant	$\geq 50\%$	colourless to pale pink
<i>Staphylococcus aureus</i> ATCC 25923	$\geq 10^3$	inhibited	0%	
<i>Enterobacter cloacae</i> ATCC 13047	50-100	good	40-50%	pink
<i>Salmonella Typhimurium</i> ATCC 14028	50-100	good-luxuriant	$\geq 50\%$	colourless
<i>Salmonella Enteritidis</i> ATCC 13076	50-100	good-luxuriant	$\geq 50\%$	colourless
<i>Shigella flexneri</i> ATCC 12022	50-100	good-luxuriant	$\geq 50\%$	colourless

**Storage and Shelf Life**

Store below 30°C in tightly closed container and prepared medium at 2-8°C and away from light to avoid photo oxidation. Use before expiry date on the label.

**Reference**

1. Endo, 1904, Zentralbl. Bakteriol., Abt. I. Orig., 35:109.
2. Eaton A. D., Clesceri L. S., Rice E. W. and Greenberg A. W., (Eds.), 2005, Standard Methods for the Examination of Water and Wastewater, 21st Ed., APHA, Washington, D.C.
3. Downes F. P. and Ito K.,(Eds.), 2001, Compendium of Methods for the Microbiological Examination of foods, 4th Ed., American Public Health Association, Washington, D.C.
4. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.

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