

EMB HiVeg™ Agar / Broth**MV317/MV503**

EMB HiVeg Agar / Broth (Eosin Methylene Blue HiVeg Agar / Broth) are recommended for the isolation and differentiation of gram negative enteric bacteria from clinical and non-clinical specimens.

Composition :**

Ingredients	MV317	MV503
	Grams/Litre	Grams/Litre
HiVeg peptone	10.00	10.00
Dipotassium phosphate	2.00	2.00
Lactose	5.00	5.00
Sucrose	5.00	5.00
Eosin - Y	0.40	0.40
Methylene blue	0.065	0.065
Agar	13.50	—

Final pH (at 25°C) 7.2 ± 0.2

** Formula adjusted, standardized to suit performance parameters

Directions :

Suspend 36.0 grams of MV317 or 22.5 grams of MV503 in 1000 ml distilled water. Mix until suspension is uniform. Heat to dissolve the medium completely. Dispense and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. AVOID OVERHEATING. Cool to 50°C and shake the medium in order to oxidize the methylene blue (i.e. to restore its blue colour) and to suspend the flocculent precipitate. (If EMB Agar is inoculated on the same day, it may be used without autoclave sterilization).

Precaution : Store the medium away from light to avoid photooxidation.

Principle and Interpretation :

These media are prepared by using HiVeg peptone in place of peptic digest of animal tissue which is free of BSE/TSE risks. EMB HiVeg Media are the modification of Eosin Methylene Blue (EMB) Media which are originally devised by Holt-Harris and Teague (1) and further modified by Levine (2).

Methylene blue and Eosin-Y inhibit gram positive bacteria to a limited degree. These dyes serve as differential indicators in response to the fermentation of carbohydrates. The ratio of eosin and methylene blue is adjusted approximately to 6:1. Sucrose is added to the medium as an alternative carbohydrate source for typically lactose-fermenting, gram-negative bacilli, which on occasion do not ferment lactose or do so slowly. The coliforms produce purplish black colonies due to taking up of methylene blue-eosin dye complex, when the pH drops. The dye complex is absorbed into the colony. Nonfermenters probably raise the pH of surrounding medium by oxidative deamination of protein, which solubilizes the methylene blue-eosin complex resulting in colourless colonies (3). HiVeg peptone serves as nitrogen source. Some strains of *Salmonella* and *Shigella* species do not grow in the presence of eosin and methylene blue. Inoculated plates should be protected from light. Further tests are required to confirm the isolates.

Product Profile :

Vegetable based (Code MV)®	Animal based (Code M)
MV317/MV503 HiVeg peptone	M317/M503 Peptic digest of animal tissue
Recommended for	: The isolation and differentiation of Gram negative enteric bacteria.
Reconstitution	: (MV317) : 36.0 g/l : (MV503) : 22.5 g/l
Quantity on preparation (500g)	: (MV317) : 13.88 L
(100g)	: (MV317) : 2.77 L
(500g)	: (MV503) : 22.22 L
(100g)	: (MV503) : 4.44 L
pH (25°C)	: 7.2 ± 0.2
Supplement	: None
Sterilization	: 121°C / 15 minutes.
Storage	: Dry Medium - Below 30°C, Prepared Medium 2 - 8°C.

Quality Control :**Appearance of Powder**

Light purple coloured, homogeneous, free flowing powder, may contain minute to small dark red purple particles.

Gelling

Firm, comparable with 1.35% Agar gel of MV317.

Colour and Clarity

Reddish-purple coloured, opalescent gel or solution having greenish cast forms in petri plates.

Reaction

Reaction of 3.6% w/v aqueous solution of MV317 and 2.25% w/v aqueous solution of MV503 is pH 7.2 ± 0.2 at 25°C.

Cultural Response

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

Organisms (ATCC)	Inoculum (CFU)	Growth	Recovery	Colour of colony (MV317)
<i>Escherichia coli</i> (25922)	10 ³ -10 ⁴	luxuriant	>70%	purple with black centre with green metallic sheen
<i>Proteus mirabilis</i> (25933)	10 ³ -10 ⁴	luxuriant	>70%	colourless
<i>Salmonella</i> serotype Typhimurium (14028)	10 ³ -10 ⁴	luxuriant	>70%	colourless
<i>Enterobacter aerogenes</i> (13048)	10 ³ -10 ⁴	good	>50%	pink, without sheen
<i>Klebsiella pneumoniae</i> (13883)	10 ³ -10 ⁴	good	>50%	pink, mucoid
<i>Staphylococcus aureus</i> (25923)	10 ⁴ -10 ⁵	inhibited	0%	—

References :

- Holt-Harris and Teague, 1916, J. Infect. Dis., 18 : 596.
- Levine, 1918, J. Infect. Dis., 23:43.
- Howard B.J., 1994, Clinical and Pathogenic Microbiology, 2nd ed., Mosby Year Book, Inc.