

Kanamycin Esculin Azide HiVeg™ Agar / Broth Base

MV510A/MV776A

Kanamycin Esculin Azide HiVeg Agar/Broth Base is used for isolation of *Enterococci* in foodstuffs.

Composition :**

Ingredients	MV510A	MV776A
	Grams/Litre	Grams/Litre
HiVeg hydrolysate	20.00	20.00
Yeast extract	5.00	5.00
Sodium chloride	5.00	5.00
Sodium citrate	1.00	1.00
Esculin	1.00	1.00
Ferric ammonium citrate	0.50	0.50
Sodium azide	0.15	0.15
Agar	10.00	-

Final pH (at 25°C) 7.0 ± 0.2

** Formula adjusted, standardized to suit performance parameters

Directions :

Suspend 21.32 grams of MV510A or 16.32grams of MV776A in 500 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45 – 50°C and aseptically add rehydrated contents of one vial of Kanamycin Sulphate Selective Supplement (FD146). Mix well before pouring into sterile petri plates/ tubes.

Warning: Sodium azide has a tendency to form explosive metal azides with plumbing materials. It is advisable to use enough water to flush off the disposables.

Principle and Interpretation :

These media are prepared by replacing Casein enzymic hydrolysate with HiVeg hydrolysate which is free from BSE/ TSE risks. Kanamycin Esculin Azide HiVeg Media are the modification of Kanamycin Esculin Azide Media which are formulated as per Mossel et al (1,2) to detect *Enterococci* in food stuffs. It is used in the dip slide technique for bacteriological monitoring of foods.

HiVeg hydrolysate, yeast extract provides essential nutrients for *Enterococci*. Kanamycin sulphate and sodium azide are the selective inhibitory components. Esculin and Ferric ammonium citrate together form indicator system to detect esculin - hydrolysing *Enterococci* forming black zones around the colonies.

The following procedure is adopted - 1gm or 1ml mixed food is added to prechilled diluent (Tryptone water HiVeg MV463) and decimal dilutions are prepared. The decimal dilution are inoculated in Kanamycin Esculin Azide HiVeg Broth Base (MV776A) and incubated at 35° for 16 -24 hours. If blackening of medium occurs, streaking is done on agar (MV510A) and after incubation confirmatory tests are carried out.

Product Profile :

Vegetable based (Code MV)©		Animal based (Code M)	
MV510A/MV776A HiVeg hydrolysate		M510A/M776A Casein enzymic hydrolysate	
Recommended for	:	Isolation of <i>Enterococci</i> in foodstuffs.	
Reconstitution	:	(MV510A) : 42.64 g/l (MV776A) : 32.64 g/l	
Quantity on preparation (100g)	:	(MV510A) : 2.34 L (100g) : (MV776A) : 3.06 L	
pH (25°C)	:	7.0 ± 0.2	
Supplement	:	Kanamycin Sulphate Selective Supplement (FD146)	
Sterilization	:	121°C / 15 minutes.	
Storage	:	Dry Medium - Below 30°C, Prepared Medium 2 - 8°C.	

Quality Control:**Appearance of Powder**

Light yellow coloured, may have slightly greenish tinge, homogeneous, free flowing powder.

Gelling

Firm, comparable with 1.0% Agar gel of MV510A.

Colour and Clarity

Medium amber coloured, clear to slightly opalescent gel forms in petri plates, clear solution in tubes.

Reaction

Reaction of 4.26% w/v of M510A or 3.27% w/v of M776A aqueous solution is pH 7.0 ± 0.2 at 25°C.

Cultural Response

Cultural characteristics observed after an incubation at 35°C or 42°C for 18-24 hours.

Organism (ATCC)	Inoculum (CFU)	Growth*	Recovery	Esculin hydrolysis
<i>Escherichia coli</i> (25922)	>10 ⁵	inhibited	0%	-
<i>Enterococcus faecium</i> (19434)	10 ³ -10 ⁵	luxuriant	>70%	+
<i>Enterococcus bovis</i> (27960)	10 ³ -10 ⁵	luxuriant	>70%	+
<i>Staphylococcus aureus</i> (25923)	10 ³ -10 ⁵	inhibited	0%	-
<i>Enterococcus faecalis</i> (29212)	10 ³ -10 ³	luxuriant	>70%	+

KEY : * = with addition of Kanamycin Sulphate Selective Supplement (FD146).

+ = Blackening of the medium / black zone around the colony.

- = No blackening of the medium / no black zone around the colony.

References :

- Mossel D.A.A., Bijker, P.G.H. and Eelderink I., 1978, Arch. Lebensmittel - Hyg., 29:121.
- Mossel D.A.A., et al., 1978, In : Streptococci., Skinner F.A. and Quesnel L. B. (ed.), SAB Symposium, series No.7, Academic Press, London.