Nutrient HiVeg™ Agar w/ Manganese MV931

Nutrient HiVeg Agar with Manganese is used for promoting sporulation of aerobic sporeformers particularly *Bacillus* species and to primarily differentiate mesophilic from thermophilic *Bacillus* species.

Composition ** :

Ingredients	Grams/Litre
HiVeg extract	3.0
HiVeg peptone No. 2	5.0
Manganese sulphate	0.03
Agar	15.0

Final pH (at 25°C) 6.8 ± 0.2
** Formula adjusted, standardized to suit performance parameters.

Directions :

Suspend 23.03 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Principle and Interpretation :

Nutrient HiVeg Agar with Manganese is prepared by using HiVeg peptone No.2 and HiVeg extract, veg substitutes for Pancreatic digest of gelatin and Beef extract respectively which are free from BSE/TSE risks normally associated with animal based peptones. Nutrient HiVeg Agar w/Manganese can be used against animal based Nutrient Agar w/ Manganese, conventionally abbreviated as NAMn which favours culture and sporulation of aerobic *Bacillus* species especially from canned foods.
HiVeg extract and HiVeg peptone No.2 provides necessary nutrients for growth of *Bacillus* species. Manganese is known to influence and enhance sporulation in the *Bacillus* species (1,2,3,4). It has been reported that organisms recovered from spoilage of foods such as fruit drinks, tomatoes, acidified onions and other canned foods sporulate well aerobically on Nutrient Agar with added manganese (5).
Thermophilic bacteria such as *Bacillus stearothermophilus* are capable of growth at 55 - 65°C while an incubation period of 30 to 35°C is favourable for culture and sporulation of mesophilic spore formers (5). This property is exploited to grow and therefore differentiate mesophilic and thermophilic spoilage bacteria. As recommended by APHA, in routine diagnosis for spoilage in canned foods, microbiological cultural procedures involves the use of primary recovery media and subculture media to identify spoilage bacteria and study their growth characteristics. Recovery medias for aerobes generally include DTA (Dextrose Tryptone Agar, M092) or DTB (Dextrose Tryptone Broth, M122). Use of Cooked meat medium (M149) is recommended for recovery of anaerobic organisms. NAMn is widely used as subculture media for aerobes.

Product Profile :

Vegetable based (Code MV)◉	Animal based (Code M)
MV931 HiVeg peptone No.2 Hiveg extract	**M931** Pancreatic digest of gelatin Beef extract

Recommended for	:	Promoting sporulation of aerobic sporeformers, particularly *Bacillus* species and to primarily differentiate mesophilic from thermophilic *Bacillus* species.
Reconstitution	:	23.03 g/l
Quantity on preparation (500g)	:	21.71 L
pH (25°C)	:	6.8 ± 0.2
Supplement	:	None
Sterilization	:	121°C / 15 minutes.

Storage : Dry Medium - Below 30°C, Prepared Medium 2 - 8°C.

Nutrient HiVeg Agar with Manganese, like the conventional medium supports growth and enhances spore production by aerobic sporeformers and therefore serves the same purpose as NAMn. When rod shaped aerobes in pure culture are isolated on HiVeg DTA (or DTB) medias (MV092/ MV122) and sporulation is not evident, the isolates should be subcultured on Nutrient HiVeg Agar with Manganese, at the temperature of initial isolation. After incubation upto 10 days, if spore production has taken place, the spores are heat shocked to destroy all vegetative cells and cultured again on HiVeg NAMn at both 30 to 35°C and 55°C. The temperature at which outgrowth occurs from the spore state indicates whether the isolate is an obligate mesophile (growth at 30 to 35°C), an obligate thermophile (growth at 55°C) or a facultative thermophile (growth at 30° to 35°C and at 55°C).

Quality Control :

Appearance of powder
Yellow coloured, may have slightly greenish tinge, homogeneous, free flowing powder.
Gelling
Firm, comparable with 1.5% Agar gel.
Colour and Clarity
Light amber coloured, clear to slightly opalescent gel forms in petri plates.
Reaction
Reaction of 2.3% w/v aqueous solution is pH 6.8 ± 0.2 at 25°C.

Continued ...

◉ Prepared from GMO free Vegetable proteins replacing Animal based peptones. Freedom from BSE/TSE worries.

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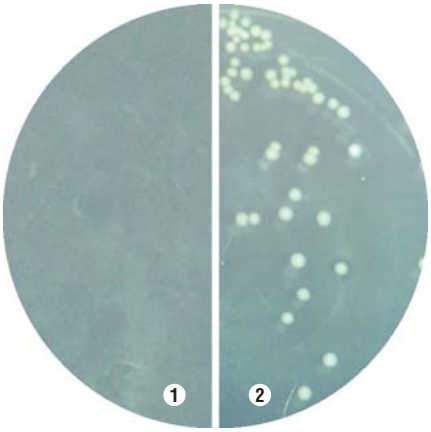

MV931 Nutrient HiVeg Agar w/ Manganese
(Against dark background)

1. Control
2. *Bacillus subtilis*

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for up to 5 days.

Organisms (ATCC)	Inoculum (CFU)	Growth	Recovery
**Bacillus stearothermophilus* (7953)	10^2-10^3	luxuriant	>70%
Bacillus coagulans (8038)	10^2-10^3	luxuriant**	>70%
Bacillus licheniformis (9945a)	10^2-10^3	luxuriant**	>70%
Bacillus megaterium (9855)	10^2-10^3	luxuriant**	>70%
Bacillus polymyxa (8526)	10^2-10^3	luxuriant**	>70%
Bacillus subtilis (6633)	10^2-10^3	luxuriant**	>70%

Key : * = Incubation at 55°C upto 7 days
**= with sporulation

References :

1. Charney, J., Fisher, W.P. and Hegarty, C.P. 1951. Manganese as an essential element for sporulation in the genus Bacilllus. J. Bacteriol. 62:145.
2. Curran, H.R. and Evans, F.R. 1954. The influence of iron or manganese upon the formation of spores by mesophilic aerobes in fluid organic media. J. Bacteriol. 67:489.
3. Maunder, D.T. 1970. "Examination of canned foods for microbial spoilage." Microbiology, Metal Div. R. and D, Continental Can Co., Inc., Oak Brook, III.
4. Penna, T.C., Machoshvili, I.A., Taqueda, M.E and Ferraz, C.A. 1998. PDA J. Pharm. Sci.Technol., 52 (5):198.
5. Downes FP and Ito K (Eds.), 2001, Compendium of methods for the microbiological examination of foods, 4th ed., APHA, Washington, D.C.