



Antibiotic HiVeg Assay Medium No. 6

MV223

Antibiotic HiVeg Assay Medium No. 6 is used for induction of spore production in *Bacillus subtilis* strains used in antibiotic assay.

Composition**

Ingredients	Gms / Litre
HiVeg hydrolysate	17.000
Papaic digest of soyabean meal	3.000
Sodium chloride	5.000
Dextrose	2.500
Dipotassium phosphate	2.500
Manganese sulphate	0.030
Final pH (at 25°C)	7.0±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 30.03 grams in 1000 ml purified/distilled water. Heat if necessary to dissolve the medium completely. Dispense in tubes and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Principle And Interpretation

Antibiotic HiVeg Assay Medium No. 6 is prepared by incorporating vegetable peptones in place of animal peptones, making the medium BSE-TSE risks free. This can be used for the same purpose of Antibiotic Assay Medium No. 6, a common medium for the assay of various antibiotics. Antibiotic Assay media are used in the performance of antibiotic assays. Grove and Randall have elaborately elucidated the methods to perform these assays and various media used for that (1). Schmidt and Moyer have reported the use of antibiotic assay medium for the liquid formulation used in the performance of antibiotic assay (2). These media are recommended by USP (3) and FDA (4). This broth is a modification of animal based Antibiotic Assay Medium No. 6 and is also used for sterility checking procedures of several preparations. It can be used for inoculum development and spore induction of *Bacillus subtilis* for antibiotic assay. Manganese is known to influence and enhance sporulation in the *Bacillus* species (5, 6). 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 (7). 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 (8).

HiVeg hydrolysate and papaic digest of soyabean meal provides the nutrients and growth factors. Dextrose provides as energy source. Dipotassium phosphate provides the buffering system. Manganese sulphate helps in the early initiation of *Bacillus* species.

Note: For Antibiotic Assay Methods and Selection of Antibiotic HiVeg Assay Medias Refer Section Antibiotic HiVeg Assay Media.

Quality Control

Appearance

Cream to yellow Homogeneous Free flowing powder

Colour and Clarity

Light amber clear may contain a slight precipitate.

Reaction

Reaction of 3.0% w/v aqueous solution at 25°C. pH : 7.0±0.2

pH

6.80-7.20

Cultural Response

MV223: Cultural characteristics observed after an incubation at different temperatures for 6 days.

Organism	Inoculum (CFU)	Growth	Incubated at	Spores
<i>Bacillus cereus</i> ATCC 10876	50-100	luxuriant	30°C	Positive
<i>Bacillus stearothermophilus</i> ATCC 25611	50-100	luxuriant	55°C	Positive
<i>Bacillus subtilis</i> ATCC 6633	50-100	luxuriant	35°C	Positive
<i>Bacillus pumilus</i> ATCC 14884	50-100	luxuriant	35°C	Positive

Storage and Shelf Life

Store below 30°C in tightly closed container and use freshly prepared medium. Use before expiry date on the label.

Reference

1. Grove and Randall, Assay Methods of Antibiotics Medical Encyclopedia, Inc. New York. .
2. Schmidt and Moyer JB, 47:199.
3. United States Pharmacopoeia 2011 , USP 34/NF 29 , US Pharmacopoeial Convention, Inc., Rockville, MD.
4. Tests and Methods of Assay of Antibiotics and Antibiotic containing Drugs F, CFR, 1983. Title 21, part 436, Subpart D, Washington, D.C. U.S Government printing office, paragraphs 436, 100-436, 106 pg 242-259 (April 1).
5. Vasantha and Freese, J.Gen.Microbiol. 112:329-336.
6. Charney J, Fisher, W.P. and Hegarty, C.P. 1951. J. Bacteriol. 62:1.
7. Downes FP& Itok (EDs) 2001.Compendium of methods for the microbiological examination of foods. 4th ed.APHA,Washington, DC.
8. Maunder DTEocffinsM, Metal Div. R. and D, Continental Can Co., Inc., Oak Brook, III.

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