Triple Sugar Iron HiVeg™ Agar MV021

Triple Sugar Iron HiVeg Agar is used for the differentiation of gram negative enteric bacilli on the basis of dextrose, lactose and sucrose fermentation and hydrogen sulphide production.

Composition ** :

Ingredients	Grams/Litre
HiVeg peptone	10.0
HiVeg hydrolysate	10.0
Yeast extract	3.0
HiVeg extract	3.0
Lactose	10.0
Sucrose	10.0
Dextrose	1.0
Sodium chloride	5.0
Ferrous sulphate	0.2
Sodium thiosulphate	0.3
Phenol red	0.024
Agar	12.0

Final pH (at 25°C) 7.4 ± 0.2
** Formula adjusted, standardized to suit performance parameters.

Directions :

Suspend 65 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Mix well and distribute into test tubes. Sterilize by autoclaving at 10 lbs pressure (115°C) for 15 minutes. Allow the medium to set in sloped form with a butt about 1 inch long.

Principle and Interpretation :

Triple Sugar Iron HiVeg Agar is prepared by completely replacing animal based peptone with vegetable peptones, making the medium free of BSE/TSE risks. Triple Sugar Iron HiVeg Agar is the modification of Triple Sugar Iron Agar which was originally proposed by Sulkin and Willett (1) and modified by Hajna (2) for identifying *Enterobacteriaceae*. Triple Sugar Iron HiVeg Agar is equivalent in performance with animal based medium which is recommended by APHA for the examination of meat and food products (3), for the examination of milk and dairy products (4) and for microbial limit test for confirming the presence of *Salmonellae* (5, 6) and in the identification of gram-negative bacilli (6, 7).
HiVeg hydrolysate, HiVeg peptone, yeast extract and HiVeg extract provide nitrogenous compounds, trace elements and vitamin B complex etc. Sodium chloride maintains osmotic equilibrium. Lactose, sucrose and dextrose are the fermentable carbohydrates. Sodium thiosulphate and ferrous ions make H_2S (hydrogen sulphide) indicator system. Phenol red is the pH indicator. Organisms that ferment glucose produce a variety of acids, changing the colour of the medium from red to yellow. More amount of acids are liberated in butt (fermentation) than in the slant (respiration). Growing bacteria also form alkaline products from the oxidative decarboxylation of peptone and these alkaline products neutralize the large amounts of acid present in the butt. Thus the appearance of an alkaline (red) slant and an acid (yellow) butt after incubation indicates that the organism is a glucose fermenter but is unable to ferment lactose and/or sucrose. To enhance the alkaline condition of the slant, free exchange of air must be permitted by closing the tube cap loosely. If the tube is tightly closed, an acid reaction caused solely by dextrose fermentation will also involve the slant. Bacteria that ferment lactose or sucrose (or both), in addition to glucose, produce large amounts of acid enables no reversion of pH in that region and thus bacteria exhibit an acid slant and acid butt. Thiosulphate is reduced to hydrogen sulphide by several species of bacteria and H_2S combines with ferric ions of ferric salts to produce the insoluble black precipitate of ferrous sulphide. Reduction of thiosulphate proceeds only in an acid environment and blackening usually occurs in the butt of the tube. Some members of the *Enterobacteriaceae* and H_2S producing *Salmonella* may not be H_2S positive on TSI HiVeg Agar. Some bacteria may show H_2S production on Kligler Iron HiVeg Agar but not on TSI HiVeg Agar. This can happen because utilization of sucrose in TSI HiVeg Agar suppresses the enzymic pathway that result in H_2S production.
Gas production (CO_2) is detected by the presence of cracks or bubbles in the medium, when the accumulated gas escapes. Do not use an inoculating loop to inoculate a tube of TSI, while stabbing the butt, mechanical splitting of the medium occurs, causing a false positive result for gas production . Triple Sugar Iron HiVeg Agar should be used in parallel with Urea HiVeg Agar/ Broth (MV112/M111) to distinguish between *Salmonella* and *Proteus* species.

Product Profile :

Vegetable based (Code MV)◉	Animal based (Code M)
MV021 HiVeg peptone HiVeg hydrolysate HiVeg extract	**M021** Peptic digest of animal tissue Casein enzymic hydrolysate Beef extract

Recommended for	:	Differentiation of enteric pathogens on the basis of dextrose, lactose, sucrose fermentation and hydrogen sulphide production.
Reconstitution	:	65.0 g/l
Quantity on preparation (500g)	:	7.69 L
(100g)	:	1.53 L
pH (25°C)	:	7.4 ± 0.2
Supplement	:	None
Sterilization	:	115°C / 15 minutes.

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

Continued ...

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

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The reactions can be summarized as follows:

Alkaline slant / acid butt	- only glucose fermented
Acid slant / acid butt	- glucose and sucrose fermented or glucose and lactose fermented or all the three sugars, glucose, lactose and sucrose fermented.
Bubbles or cracks present	- gas production
Black precipitate present	- H_2S gas production.

Quality Control :

Appearance of powder

Beige to light pink coloured, homogeneous, free flowing powder.

Gelling

Firm, comparable with 1.2% Agar gel.

Colour and Clarity

Pinkish red coloured, clear to slightly opalescent gel forms in tubes as slants.

Reaction

Reaction of 6.5% w/v aqueous solution is pH 7.4 ± 0.2 at 25°C.

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organisms (ATCC)	Growth	Slant	Butt	Gas	H_2S
Citrobacter freundii (8090)	luxuriant	A	A	+	+
Enterobacter aerogenes (13048)	luxuriant	A	A	+	-
Escherichia coli (25922)	luxuriant	A	A	+	-
Klebsiella pneumoniae (13883)	luxuriant	A	A	+	-
Proteus vulgaris (13315)	luxuriant	K	A	-	+
Salmonella serotype Paratyphi A	luxuriant	K	A	+	-
Salmonella serotype Typhi (6539)	luxuriant	K	A	-	+
Salmonella serotype Typhimurium (14028)	luxuriant	K	A	+	+
Shigella flexneri (12022)	luxuriant	K	A	-	-

Key : A = acidic, yellow
K = alkaline, no change
+ = positive reaction H_2S / gas production
- = no reaction no H_2S / gas production

References :

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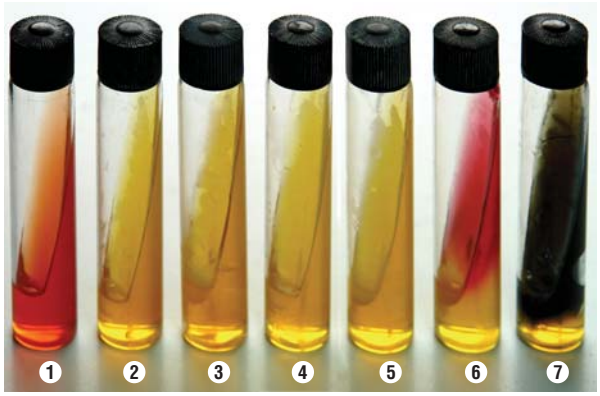
3. Downes FP and Ito K (Eds.), 2001, Compendium of Methods For The Microbiological Examination of Foods, 4th ed., APHA, Washington, D.C.
4. Standard Methods for the Examination of Dairy Products. 17th Edition, 2004 Edited by H. Michael Wehr and Joseph H.Frank.
5. Forbes BA, Sahm DF, Weissfeld AS, 2002, Bailey and Scott's Diagnostic Microbiology, 11th ed., The C.V. Mosby Co., St. Louis.
6. Eaton A.D., Clesceri L.S. and Greenberg A.E., (Eds.), 2005, Standard Methods for the Examination of Water and Wastewater, 21st ed, APHA, Washington DC.
7. MacFaddin J., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore.



MV021 Triple Sugar Iron HiVeg Agar

1. Control
2. *Citrobacter freundii*
3. *Enterobacter aerogenes*
4. *Escherichia coli*
5. *Klebsiella pneumoniae*
6. *Salmonella* serotype Typhi
7. *Salmonella* serotype Typhimurium