

**DNase Test HiVeg™ Agar Base****MV482**

DNase Test HiVeg Agar is recommended for the detection of deoxyribonuclease activity of bacteria and fungi, and especially for identification of pathogenic *Staphylococci*.

**Composition \*\* :**

Ingredients	Grams/Litre
HiVeg hydrolysate	15.0
Papaic digest of soyabean meal	5.0
Deoxyribonucleic acid (DNA)	2.0
Sodium chloride	5.0
Agar	15.0

Final pH (at 25°C ) 7.3 ± 0.2

\*\* Formula adjusted, standardized to suit performance parameters.

**Directions :**

Suspend 42 grams in 1000 ml distilled water. Heat to boiling with frequent agitation to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45°C and pour into sterile petriplates. Add rehydrated contents of one vial of Toluidine Blue (FD051) before sterilizing the medium or flood the plates with 0.1% Toluidine Blue (FD051) after incubation.

**Principle and Interpretation :**

DNase Test HiVeg Agar Base is specially developed from HiVeg hydrolysate to avoid BSE/TSE risks associated with animal origin casein enzymic hydrolysate. DNase Test HiVeg Agar is the modification of DNase Test Agar which is used for detecting deoxyribonuclease activity of bacteria and fungi and particularly for identification of pathogenic *Staphylococci*. With added toluidine blue, it is used in differentiation and identification of nonpigmented *Serratia* species isolated from clinical sources that might be improperly identified as *Enterobacter* and *Klebsiella* species. DNase activity was observed by Weckman and Catlin (1) in *Micrococci* and found the correlation with coagulase activity as coagulase positive species were DNase positive. DiSalvo (2) confirmed the results of Weckman and Catlin and observed accurate correlation of DNase and coagulase activity. In his experiment Di Salvo incorporated DNA and calcium chloride to activate DNase enzyme. DNase medium was modified by adding toluidine blue by Schreier (3). Modified medium achieved faster identification of *Serratia marcescens* and could differentiate *Serratia* from other members of the *Enterobacteriaceae*.

HiVeg hydrolysate, papaic digest of soyabean meal provide essential nutrients. The DNase depolymerizes the DNA resulting in the formation of a clear zone around the microbial growth which is visualized by flooding the plate with 1N hydrochloric acid (4). When toluidine blue is added to the medium itself then, DNase activity results in the production of a bright pink zones surrounding growth due to the metachromatic property of toluidine blue. Some strains of *Staphylococci* may be inhibited on DNase Test Agar due to toluidine blue. Further confirmatory tests for the identification should be carried out.

**Product Profile :**

Vegetable based (Code MV)©	Animal based (Code M)
<b>MV482</b> HiVeg hydrolysate	<b>M482</b> Casein enzymic hydrolysate

**Recommended for** : Detection of deoxyribonuclease activity of bacteria and fungi, especially for pathogenic *Staphylococci*.

**Reconstitution** : 42.0 g/l

**Quantity on preparation (100g)** : 2.38 L

**pH (25°C)** : 7.3 ± 0.2

**Supplement** : Toluidine blue (FD051)

**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 1.5% Agar gel.

**Colour and Clarity**

Light amber coloured, clear to slightly opalescent gel forms in petri plates or blue coloured, clear to slightly opalescent gel forms in petriplates when Toluidine blue (FD051) is added.

**Reaction**

Reaction of 4.2% w/v aqueous solution is pH 7.3 ± 0.2 at 25°C

**Cultural Response**

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

Organisms (ATCC)	Inoculum (CFU)	Growth	DNase Activity
<i>Serratia marcescens</i> (8100)	10 <sup>2</sup> -10 <sup>3</sup>	luxuriant	+
<i>Staphylococcus aureus</i> (25923)	10 <sup>2</sup> -10 <sup>3</sup>	luxuriant	+
<i>Staphylococcus epidermidis</i> (12228)	10 <sup>2</sup> -10 <sup>3</sup>	luxuriant	-
<i>Streptococcus pyogenes</i> (19615)	10 <sup>2</sup> -10 <sup>3</sup>	luxuriant	+

Key : On flooding the DNase Test HiVeg Agar plate with 1N HCl

+ = clear area surrounding growth

- = no clearing surrounding growth

DNase Test Agar with Toluidine blue

+ = pink to red zone surrounding growth

- = no colour change surrounding growth

**References :**

1. Weckman and Catlin, 1957, J. Bact., 73:747.
2. Di Salvo, 1958, Med. Tech. Bull., U.S. Armed Forces Med. J., 9:191.
3. Schreier, 1969, Am. J. Clin. Pathol., 51:711.
4. Streifeld, Hoffman and Janklow, 1962, J.Bact., 84:77.