



# **HiCrome Nickels and Leesment HiVeg Medium**

**MV1712** 

HiCrome Nickels and Leesment HiVeg Medium is used for the enumeration of citrate-fermenting lactic acid bacteria from milk, milk products and mesophilic starter cultures.

# **Composition\*\***

Ingredients	Gms / Litre
HiVeg hydrolysate	18.000
Yeast extract	4.500
Gelatine	2.250
Glucose	4.500
Lactose	4.500
Sodium chloride	3.600
Trisodium citrate dihydrate	1.800
Calcium lactate pentahydrate	8.000
Tricalcium dicitrate, dihydrate	6.650
Carboxymethyl cellulose (CMC)	0.400
Chromogenic substrate (X-gal)	0.200
Agar	15.000
Final pH ( at 25°C)	$6.65 \pm 0.05$
**Formula adjusted standardized to suit performance parameters	

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

# Directions

Suspend 66.0 grams (the equivalent weight of dehydrated medium per litre) in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15lbs pressure (121°C) for 15 minutes. Cool to 45-50°C. If desired, add rehydrated contents of 2 vials of HiCrome Nickels and Leesment Selective Supplement (FD245). Mix well and pour into sterile Petri plates.

# **Principle And Interpretation**

Lactic acid bacteria are widespread in nature and are best known for their activities in major food such as dairy, meat and vegetable products (1).

Testing for lactic acid bacteria in dairy products may be useful for various reasons like evaluating lactic starter cultures; determining the cause of acid defects in milk products, controlling the quality of cured cheese, cultured milks and uncultured products containing added cultures (2).

HiCrome Nickels and Leesment HiVeg Medium is a modification of HiCrome Nickels and Leesment Medium wherein casein enzymic hydrolysate is replaced by HiVeg hydrolysate to avoid BSE/TSE risks associated with animal origin peptone. The animal version is a modification of Modified Nickels and Leesment Medium formulated as per APHA (1) and is used for the enumeration of citrate-fermenting lactic acid bacteria using colony count technique at 25°C.

HiVeg hydrolysate and yeast extract serve as carbon and nitrogen sources. Lactose and glucose are the carbohydrate source in the medium. X-gal differentiates between *Lactococcus lactis* subsp. *lactis* and *Leuconostoc* species. Citrate in the medium is solubilized by *Lactococcus lactis* subsp. *lactis biovar diacetylactis* and thus gives clear zones around the colony.

*Lactococcus lactis* subsp. *lactis biovar diacetylactis* colonies are white with a clear zone. *Lactococcus lactis* subsp. *lactis* and *Lactococcus lactis* subsp. *cremoris* colonies are white without a clear zone. *Leuconostoc* species are blue, with or without a clear zone. HiCrome Nickels and Leesment Medium with the addition of HiCrome Nickels and Leesment Supplement (FD245) can be used for enumeration of *Leuconostoc* (1). Vancomycin acts as a supplement for the selective isolation of *Leuconostoc* from a mix flora of lactic acid bacteria. Sodium chloride maintains osmotic equilibrium and various salts provides essential ions.

Cream to light yellow homogeneous free flowing powder

#### Gelling

Firm, comparable with 1.5% Agar gel

### Colour and Clarity of prepared medium

White coloured, opaque gel containing white precipitate forms in Petri plates.

#### Reaction

Reaction of 6.6% w/v aqueous solution at 25°C. pH : 6.65±0.05

#### pН

6.60-6.70

### **Cultural Response**

Cultural characteristics observed after an incubation at 25-30°C for 48-72 hours.

#### **Cultural Response**

Organism	Growth	Growth with FD245	Colour of colony
Cultural Response			
L. lactis biovar diacetylactis	good-luxuriant	inhibited	white with a clear zone
L. lactis subsp lactis ATCC 19435	good-luxuriant	inhibited	white without a clear zone
L. lactis subsp cremoris ATCC 19257	good-luxuriant	inhibited	white without a clear zone
<i>Leuconostoc mesenteroides</i> <i>ATCC 9135</i>	good-luxuriant	good-luxuriant	blue without clear zone

# **Storage and Shelf Life**

Store dehydrated powder and prepared medium at 2-8°C. Use before expiry period on the label.

## Reference

1.Downes F.P. and Ito K., (Eds) 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., APHA Washington D.C.

2.Marshall R.T., 1992, Standard Methods for the Examination of Dairy products, 16th Ed, American Public Health Association, Washington D.C.

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HiMedia Laboratories Pvt. Ltd. A-516,Swastik Disha Business Park,Via Vadhani Ind. Est., LBS Marg, Mumbai-400086, India. Customer care No.: 022-6147 1919 Email: techhelp@himedialabs.com Website: www.himedialabs.com

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