BYE HiVeg™ Agar

MV470

Intended Use:

Recommended for cultivation and routine studies of distribution of Mycoplasmas or Pleuropneumonia like organisms (PPLOs) and L-forms of bacteria.

Composition**

Ingredients	g / L
HiVeg™ peptone No. 3	10.000
HiVeg™ special infusion	7.500
HiVeg™ infusion	10.000
Dextrose (Glucose)	2.000
Sodium chloride	5.000
Disodium hydrogen phosphate	2.500
Yeast extract	2.000
Agar	13.000
Final pH (at 25°C)	7.9±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 52.0 grams in 850 ml purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C and aseptically add 150 ml of sterile human or animal blood or serum. Mix gently and pour into sterile Petri plates.

Principle And Interpretation

Mycoplasmas (mollicutes) are the smallest free-living microorganisms (1). Earlier *Mycoplasmataceae* were given the general title of pleuropneumonia like organism (PPLO), because of similarities to *Mycoplasma mycoides* (subsp. mycoides), the causative agent of bovine pleuropneumonia (2). BYE media are simple media developed for cultivation and routine studies of distribution, habitat and possible pathogenesis of Mycoplasma - Pleuropneumonia like organisms and L-forms of bacteria by Barile, Yaguchi and Eveland (1). These media can be used for isolation of PPLOs from urethritis, penile ulcerations and cervical specimens and L-forms of *Corynebacterium*, *Neisseria*, and *Streptococcus*. These are also used for detecting PPLO contamination of tissue culture and cell-lines (3) and for membrane filter work (4).

BYE HiVeg™ Agar is prepared by completely replacing animal based peptone with vegetable peptones to avoid BSE/TSE risks associate with animal peptones. BYE Agar contains HiVeg™ special infusion and HiVeg™ infusion along with yeast extract, which provide carbon, nitrogen, vitamins and other growth factors required for the metabolism of Mycoplasma - Pleuropneumonia like organisms. Inoculations are made in duplicates. One set is incubated aerobically while the other anaerobically for 48 hours or more. Usually growth occurs within 3-5 days; however, negative results are reported after 10 days. Anaerobic conditions are most important for the first 3 days while secondary transfers can be incubated aerobically.

Type of specimen

Clinical samples - Swabs of Sputum,Tissue, Bronchial lavage (BAL) fluid, Bronchial washings, Cerebrospinal fluid (CSF)

Specimen Collection and Handling:

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations :

1. Some organism may show poor growth due to nutritional variation.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance
Cream to yellow homogeneous free flowing powder
Gelling
Firm,comparable with 1.3% Agar gel.
Colour and Clarity of prepared medium
Yellow coloured, clear to slightly opalescent gel forms in Petri plates
Reaction
Reaction of 5.2% w/v aqueous solution at 25°C. pH : 7.9±0.2
pH
7.70-8.10
Cultural Response
Cultural characteristics observed with added serum under humidified anaerobic conditions,after an incubation at 35-37° C for 5-10 days.

Organism	Growth
Mycoplasma bovis ATCC 25523	good-luxuriant
Mycoplasma gallinarium ATCC 19708	good-luxuriant
Mycoplasma pneumoniae ATCC 15531	good-luxuriant
Streptococcus pneumoniae ATCC 6303	good-luxuriant

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (5,6).

Reference

1.Murray P.R., Baron E. J., Pfaller M.A., Tenover F.C., Yolken R.H.(Eds.),1995, Manual of Clinical Microbiology, 6th Ed., ASM Press.
2.Barile, Yaguchi, Eveland, 1958, Am. J. Clin. Path. 30:171.
3.Collee J.G, Fraser A.G., Marmion B.P., Simmons. A (Eds.), 1996, Mackie and McCartney Practical Medical Microbiology, 14th Ed, Churchill Livingstone.
4.Barile, 1962, J. Bacteriol., 83:430.
5.Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
6.Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

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