



PPLO Modified Broth Base w/o CV

M1586

PPLO Modified Broth Base w/o CV is used for isolation and cultivation of *Mycoplasma* species (pleuropneumonia like organisms).

Composition**

Ingredients	Gms / Litre
Meat infusion from	50.000
Peptone	10.000
Sodium chloride	5.000
Final pH (at 25°C)	7.8±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 21 grams in 700 ml 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 300 ml Horse serum (RM1239) or 10 vials of Mycoplasma Enrichment Supplement (FD075). Mix well before dispensing.

Principle And Interpretation

PPLO Agar was described by Morton, Smith and Leberman (1). It was used in a study of the growth requirements of *Mycoplasma* (2), along with the identification and cultivation of this organism. (3-5). Pivotal information regarding *Mycoplasma* has been documented by Sabin (6). Hayflick et al have reported the information regarding the cultivation of *Mycoplasma* (7).

PPLO Broth without crystal violet is prepared according to the formula described by Morton and Lecce (2). Crystal violet is omitted from this formula due to its inhibitory action on some *Mycoplasma*. It has been used for the cultivation of *Mycoplasma* for research studies (8, 9).

For the cultivation of *Mycoplasma* the medium ingredients and all the supplements should be free of any toxic substances even in small amounts. Many *Mycoplasma* require serum for their good growth and also presence of antibiotic is necessary to prevent the growth of contaminating organisms. Mostly the *Mycoplasma* species are aerobic or facultatively anaerobic but some are microaerophilic. Few are anaerobic saprophytic *Mycoplasma* which grow best at 22-35°C while pathogenic strains grow at 35°C.

Tubes should be incubated in an atmosphere containing 5-10% carbon dioxide and examined after incubation of 48 hours but they should not be discarded as negative until after incubation for 3 weeks.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Yellow coloured clear solution in tubes

Reaction

Reaction of 2.1% w/v aqueous solution at 25°C. pH : 7.8±0.2

pH

7.60-8.00

Cultural Response

M1586: Cultural characteristics observed in presence of 10% CO₂ with added Horse serum (RM1239) OR 10 vials of Mycoplasma Enrichment Supplement (FD075), after an incubation at 22-35°C for 24-48 hours

Organism

Growth

<i>Mycoplasma bovis</i> ATCC 25523	good-luxuriant
<i>Mycoplasma gallinarium</i> ATCC 19708	good-luxuriant
<i>Mycoplasma pneumoniae</i> ATCC 15531	good-luxuriant
<i>Streptococcus pneumoniae</i> ATCC 6303	good-luxuriant

Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label.

Reference

1. Morton, Smith and Leberman, 1951, Am. J. Syphilis Gonorrh. Venereal Diseases, 35: 361.
2. Morton and Lecce, 1953. J. Bacteriol., 66:646.
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5. Gregory and Cundy, 1970, Appl. Microbiol., 19:268.
6. Sabin, 1941, Bacteriol. Rev., 5:1, 331.
7. Hayflick and Chanock, 1965, Bacteriol, Rev., 29:185.
8. Adler and Da Massa, 1967, Appl. Microbiol., 15:245.
9. Leland, Lapworth, Jones and French, 1982, J. Clin. Microbiol., 16:709.

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