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PPLO AGAR BASE

When supplemented with an appropriate enrichment, this medium is recommended for the isolation and cultivation of Mycoplasma species, also known as pleuropneumonia-like organisms (PPLO).

REF: BS.1/DB01.100.0100	100 Gram	REF: BS.1/DB01.250.0250	250 Gram
REF: BS.1/DB01.500.0500	500 Gram		

CLINICAL SIGNIFICANCE

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 (3,4,5). Pivotal information regarding Mycoplasma has been documented by Sabin (6). Hayflick et al have reported the information regarding the cultivation of Mycoplasma (7). For the cultivation of Mycoplasma the medium ingredients and all the supplements should be free of any toxic substances even in small amounts. HM infusion B from and peptone provide nitrogen and carbon source, long chain amino acids, vitamins, and other essential nutrients. Sodium chloride maintains the osmotic balance of these formulations. 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. Mycoplasma when grow in the agar medium show typical morphology and form colonies below the agar surface and do no grow without serum. Plates or 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. PPLO colonies are round with a dense center and a less dense periphery, resembling a “fried egg” on PPLO Agar. Vacuoles, large bodies characteristic of Mycoplasma species are seen in the periphery. Colonies vary in diameter from 10 to 500 microns (0.01-0.5 mm) and penetrate into the medium.

MEDIA COMPOSITION

Item	Formula per liter of medium g/l
- Beef Heart Infusion	6.0
- Peptone	10.0
- Dextrose	5.0
- Sodium chloride	0.25
- Agar	15

pH 7.8 ± 0.2 @ 25°C

PRECAUTIONS AND WARNINGS

Media to be handled by entitled and professionally educated person. Do not ingest or inhale.

Good Laboratories practices using appropriate precautions should be followed in:

- Wearing personnel protective equipment (overall, gloves, glasses,).
- Do not pipette by mouth.
- In case of contact with eyes or skin; rinse immediately with plenty of soap and water. In case of severe injuries; seek medical advice immediately.
- Respect country requirement for waste disposal.

S56: dispose of this material and its container at hazardous or special waste collection point.
S57: use appropriate container to avoid environmental Contamination.
S61: avoid release in environment.
For further information, refer to the PPLO AGAR BASE material safety data sheet.

STORAGE AND STABILITY

BioScien PPLO AGAR BASE dehydrated media are stable until expiration date stated on label when properly stored 10-30°C. The prepared medium should be stored 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 avoid lump development due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in a 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.

MEDIA PREPARATION

Suspend 36.0 grams in 700 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 300 ml Horse serum (RM1239) or 10 vials of HPYT Enrichment Supplement (FD075). Mix well and pour into sterile Petri plates. 25% Ascitic fluid can be used instead of Horse serum.

Deterioration

The color of BioScien PPLO AGAR BASE is Cream to yellow homogeneous free flowing powder. Colour and Clarity of prepared medium IS Yellow coloured clear to slightly opalescent gel forms in Petri plates If there are any physical changes, discard the medium The hydrated medium is Amber coloured clear solution without any precipitate, media should not be used if there are any signs of deterioration (shrinking, cracking, or discoloration), and contaminations.

EQUIPMENT REQUIRED NOT PROVIDED

- Sterile cups
- Sterile tubes
- Sterile loops
- Incubator

PERFORMANCE CHARACTERISTICS

Cultural characteristics observed in presence of 10% Carbon dioxide with added,1% Horse serum (RM1239) or 10 vials ofHPYT Enrichment Supplement (FD075), after an incubation at 22-35°C for 48 hours

Test Organisms	Growth
Mycoplasma bovis ATCC 25523	Luxuriant
Mycoplasma gallinarium ATCC 19708	Luxuriant
Mycoplasma pneumoniae ATCC 15531	Luxuriant
Streptococcus pneumoniae ATCC 6303	Luxuriant

SYMBOLS IN PRODUCT LABELLING	
IVD For in-vitro diagnostic use	Number of <n> test in the pack
LOT Batch Code/Lot number	Caution
REF Catalogue Number	Do not use if package is damaged
Temperature Limitation	Consult Instruction for use
Expiration Date	
Manufactured by	

QUALITY CONTROL

To ensure adequate quality control, it is recommended that positive and negative control included in each run. If control values are found outside the defined range, check the system performance. If control still out of range please contact ***BioScien*** technical support.

REFERENCES

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