

Block 5, Street 9 inside The Ismailia free zone. Ismailia- Egypt

Post Code-41511

Tel.-Fax: +202 21813500/ +202 21813600 Mob.: +2 01211550941/ +2 01119225860 E-mail: sales@arenabioscientific.com Website: www.arenabioscien.com

# Mycoplasma Agar Base (PPLO Agar Base)

A selective medium for the isolation and cultivation of *Mycoplasma* Sp.

REF: BS.1/PP01.100.0100 100 Gram REF: BS.1/PP01.250.0250 250 Gram REF: BS.1/PP01.500.0500 500 Gram

#### **CLINICAL SIGNIFICANCE**

The pleuropneumonia-like organisms (PPLO) are a group of pleomorphic, filterable, fastidious microorganisms which also lack a rigid cell wall. This feature makes them naturally resistant to antibiotics that target cell wall synthesis (for example the beta-lactam antibiotics). *M. pneumonia* is an important causative agent of "walking pneumonia" and other respiratory disorders, and *M. genitalium*, which is thought to be involved in pelvic inflammatory diseases.

#### **METHOD PRINCIPLE**

This medium contains beef heart infusion and peptone to supply nutrients required for the growth of mycoplasmas like nitrogen, vitamins, amino acids and carbon. Sodium chloride maintains osmotic equilibrium. Agar acts as a solidifying agent. Normal horse serum provides a protein source. Penicillin and thallium acetate can added as inhibitors of bacteria. Yeasts and molds are inhibited by the amphotericin B contained in the medium.

#### **MEDIA COMPOSITION**

Item	Formula per liter of medium
Beef extract	6 gm
Peptone	10 gm
Sodium Chloride	5 gm
Agar	15 gm

#### 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 material safety data sheet.

## STORAGE AND STABILITY

**BioScien** PPLO Agar should be stored between 10-30°C in a firmly 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 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.

#### Final pH 7.8±0.2 at 25°C

#### **MEDIA PREPARATION**

- 1- Suspend 36 gm of the dry powder in 700 ml distilled water. Mix thoroughly. Heat with frequent stirring and boil for 1 minute until the powder is completely dissolved.
- 2- Autoclave at 121°C for 15 minutes. Cool the medium to 50-60°C.
- 3- Aseptically add 300 ml horse serum to the medium. Mix well.
- 4- Add Selective agents if desired (i.e. 0.25gm Thallium Acetate,
- 2.5mg Amphotericin B and 1,000,000 U Penicillin).

#### **PROCDURE**

Inoculate the surface of plates containing the complete medium by adding drops of liquid inoculum prepared by serial dilution of the sample, or by a swab-inoculation method. Incubate the media plates at  $35 \pm 2^{\circ}$ C for up to 28 days under a moist atmosphere containing 5-10% carbon dioxide or anaerobically if the presence of *M. buccale, M. faucium, M. orale or M. salivarium* is suspected

# Deterioration

The color of **BioScien** PPLO Agar is cream to yellow homogeneous free flowing powder. Prepared Media is light yellow in color. If there are any physical changes for powder or signs of deterioration (shrinking, cracking, or discoloration), and contaminations for hydrated media, discard the medium.

#### **SPECIMEN**

Clinical samples and pharmaceutical samples

#### **EQUIPMENT REQUIRED NOT PROVIDED**

- · Sterile Petri dishes
- Incubator
- Autoclave

#### PERFORMANCE CHARACTERISTICS

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 4 weeks. PPLO colonies are round with a dense center and a less dense margin, resembling a "fried egg" on PPLO Agar.

Microorganisms	Growth
Mycoplasma bovis ATCC 25523	luxuriant
Mycoplasma gallinarium ATCC 19708	luxuriant
Mycoplasma pneumoniae ATCC 15531	luxuriant

## **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**

- 1. Morton et al. (1951) Am. J. Syphil. Gonorrh. Vener. Dis. 35: 361.
- 2. Morton and Lecce, 1953. J. Bacteriol., 66:646
- 3. Craven, Wenzel, Calhoun, Hendley, Hamory and Gwaltney, 1976, J. Clin. Microbiol., 4:225.
- 4. Gregory and Cundy, 1970, Appl. Microbiol., 19:268.5. Isenberg (ed.). 1992. Clinical microbiology procedures handbook, vol. 1. American Society for Microbiology, Washington, D.C.
- 6. Baron, Peterson and Finegold. 1994. Bailey & Scott's diagnostic microbiology, 9th ed. Mosby-Year Book, Inc. St. Louis, Mo.
- 7- Kenny. 1985. In Lennette, Balows, Hausler and Shadomy (ed.). Manual of clinical microbiology, 4th ed. American Society for Microbiology, Washington, D.C.

SYMBOLS IN PRODUCT LABELLING			
For in-vitro diag	gnostic	Number of <n> test in the pack</n>	
LOT Batch Code/Lot	t number	Caution	
REF Catalogue Num	nber	Do not use if package is damaged	
Temperature Li	mitation	Consult Instruction for use	
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Manufactured by	′		