

# **Technical Information**

# **Martin Lewis Agar Plate**

# Product Code: PM 3085

Application: Recommended for the isolation and cultivation of Neisseria species from clinical specimens.

| Composition**                       |             |  |
|-------------------------------------|-------------|--|
| Ingredients                         | Gms / Litre |  |
| Tryptone                            | 7.500       |  |
| HM Peptone #                        | 7.500       |  |
| Dipotassium hydrogen phosphate      | 4.000       |  |
| Potassium phosphate                 | 1.000       |  |
| Corn starch                         | 1.000       |  |
| Sodium chloride                     | 5.000       |  |
| Haemoglobin (MS2022)                | 10.000      |  |
| Vitamino Growth Supplement (MS2025) | 1.0 vial    |  |
| VCAT Supplement (MS2353)            | 1.0 vial    |  |
| Agar                                | 12.000      |  |

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

#- Equivalent to Meat peptone

## Principle & Interpretation

Majority of gonococcal infections are uncomplicated lower genital tract infection caused by direct infection of the columnar epithelium of mucosal membranes. *Neisseria gonorrhoeae* is the causative agent of gonococcal infections. Most *Neisseria* strains have complex growth requirements; some strains may be exquisitely sensitive to fatty acids, necessitating the incorporation of soluble starch in the growth media (4).

Carpenter and Morton reported an improved medium to isolate Gonococci in 24 hours (2). Later on the efficiency of GC medium supplemented with hemoglobin and yeast concentrate was demonstrated for isolating gonococci (1). Subsequently Thayer and Martin Medium was developed for the primary isolation of *Neisseria gonorrhoeae* and *Neisseria meningitidis* from specimens containing mixed flora collected from throat, vagina, rectum and urethra (6,9). Thayer and Martin (9) used Vancomycin, Colistin and Nystatin. Martin and Lester (7) used an additional antibiotic Trimethoprim to make the medium selective.

Tryptone and HM Peptone supplies nitrogenous and carbonaceous compounds, long chain amino acids and other essential growth nutrients for the growth of fastidious organisms. Phosphates buffer the medium. Sodium chloride maintains the osmotic balance. For the cultivation of fastidious organisms the medium should be supplemented with essential growth factors supplied predominantly by yeast extract (MS2027). This can be replaced with a chemically defined supplement containing essential growth factors available from yeast extract in Vitamino Growth Supplement, (Twin Pack) (MS2025). X-factor needed for the growth of fastidious *Haemophilus* species is provided by hemoglobin (MS2022). Selective supplement inhibits accompanying bacteria.

### Type of specimen

Clinic samples

## Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (3,4). After use, contaminated materials must be sterilized by autoclaving before discarding.



# Warning and Precautions

In Vitro diagnostic use only. 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 clinical specimens. Safety guidelines may be referred in individual safety data sheets.

## Limitations :

- 1. Due to nutritional variations, certain strains may show poor growth.
- 2. Certain strains of Neisseria gonorrheae may be inhibited by antibiotics.
- 3. An enriched non-selective medium must be used in parallel.
- 4. Further biochemical and serological tests must be carried for confirmation.

### 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..

# Methodology

Either streak, inoculate or surface spread the test inoculum (50-100 CFU) aseptically on the plate.

# **Quality Control**

#### Appearance

Sterile Martin Lewis Agar in 90mm disposable plate with smooth surface and absence of black particles/cracks/bubbles. **Colour** Chocolate brown coloured medium

#### Quantity of medium

25 ml of medium in 90 mm disposable plate

### Cultural response

Cultural characteristics observed after an incubation at 35-37°C for 18 - 48 hours.

#### Cultural Response

Cultural characteristics observed in presence of 5-10% Carbon dioxide ( $CO_2$ ) and 70% humidity with added sterile 2% Haemoglobin (MS2022) and GC Supplement with antibiotics (MS2021), after an incubation at 35-37°C for 40-48 hours

| Organism                         | Inoculum(CFU) | Growth   | Recovery |
|----------------------------------|---------------|--|----------|
| Haemophilus influenza ATCC 19418 | 50-100        | luxuriant  | >=50%    |
| Neisseria gonorrhoeae ATCC19424  | 50-100        | good-luxuriant(with added<br>antibiotic supplements) | >=50%    |
| Neisseria meningitidis ATCC13090 | 50-100        | good-luxuriant(with added                            | >=50%    |



#### Ready Prepared Media

|                                   | antibiotic supplements) |                |       |
|-----------------------------------|-------------------------|----------------|-------|
| Streptococcus pyogenes ATCC19615  | 50-100                  | good-luxuriant | >=50% |
| Streptococcus pneumonia ATCC 6303 | 50-100                  | good-luxuriant | >=50% |

# Storage and Shelf Life

- On receipt store between 2-8°C Use before expiry date on the label.
- 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 (3,4).

# **Further Reading**

- 1. Carpenter et al, 1949, Am. J. Syphil. Gonorrh. Vener. Dis., 33:164.
- 2. Carpenter and Morton, 1947, Proc. N.Y. State Assoc. Public Hlth. Labs., 27:58.
- 3. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
- 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.
- 5. MacFaddin J., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. I, Williamsand Wilkins, Baltimore.
- 6. Martin, Billings, Hackney and Thayer, 1967, Public Hlth. Rep., 82:361.
- 7. Martin J.E. Jr. and Lester A., 1971, HSMHA Hlth. Service Rep., 86(1):30.
- 8. Murray P. R., Baron E. J., Pfaller M. A., Jorgensen J. H. and Yolken R. H., (Eds.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C
- 9. Thayer J. and Martin J.E. Jr., 1966, Public Health Rep., 81:559

#### **Disclaimer :**

- User must ensure suitability of the product(s) in their application prior to use.
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