HealthLink
800-638-2625

Product No. 1100
Revision No. 01

PRODUCT INFORMATION AND QUALITY CONTROL SHEET

Modified Thayer Martin Agar

I. INTENDED USE

Modified Thayer Martin Agar is an enriched and selective medium used for the isolation of pathogenic *Neisseria* species from specimens containing mixed microbiologic flora.

II. SUMMARY AND EXPLANATION

Thayer-Martin Selective Agar was developed for the isolation of *N. gonorrhoeae* and *N. meningitidis* from specimens containing mixed flora taken from the throat, vagina, rectum and urethra.[1,2] Thayer-Martin consists of a chocolate agar base to which vancomycin, colistin and nystatin have been added to minimize overgrowth by contaminants, suppress the growth of saprophytic *Neisseria* sp., and enhance the growth of pathogenic *Neisseria.*

Martin, et. al, modified Thayer-Martin Agar by incorporating trimethoprim to produce Modified Thayer-Martin (MTM) medium. Trimethoprim suppresses the growth of swarming *Proteus* sp. thereby increasing the number of gonococci isolated from clinical specimens.[3]

III. PRINCIPLES OF THE PROCEDURE

Modified Thayer Martin Agar consists of a chocolate GC agar base, bovine hemoglobin, and a chemically defined enrichment. The GC base provides nitrogenous nutrients, phosphate buffers, and cornstarch to neutralize toxic fatty acids. Hemoglobin provides X factor (hemin). Chemically defined enrichments provide V factor (nicotinamide adenine dinucleotide - NAD), vitamins, amino acids, coenzymes, dextrose, ferric ions and other growth factors necessary for optimal growth of pathogenic *Neisseria* species.

The antimicrobial agents vancomycin, colistin, nystatin and trimethoprim are incorporated to suppress competing normal flora organisms. Vancomycin actively inhibits gram positive organisms, colistin suppresses gram negative organisms other than *Proteus* sp., nystatin is effective against fungi and trimethoprim suppresses swarming *Proteus* species.

IV. TYPICAL FORMULA AND APPEARANCE

Appearance = opaque, chocolate brown

(Approximate formula* per liter of processed water)

Pancreatic Digest of Casein	7.5g
Selected Meat Peptone	7.5
Corn Starch	1.0
Dipotassium Phosphate	4.0
Monopotassium Phosphate	1.0
Sodium Chloride	5.0
Agar	15.0
Hemoglobin	10.0
Chocolate Enrichment Solution	10 ml
VCNT Inhibitor Solution	10 ml

*adjusted and/or supplemented to meet performance criteria.

V. PRECAUTIONS

This product is for IN VITRO diagnostic use only. Culture specimens may contain microorganisms which can be potentially infectious to the user. Strict adherence to aseptic techniques and established precautions against microbiological hazards should be followed throughout the procedure. Carefully dispose of all items which contact patient specimens or isolated bacteria. (See Material Safety Data Sheet for further information.)

VI. STORAGE/SHELF LIFE

Plated media should be stored at 2-8°C (36-46°F), media side up, in the unopened or resealed package protected from light. DO NOT FREEZE OR EXPOSE TO HIGH TEMPERATURES. Allow unopened plates to warm to room temperature prior to inoculation. Prior to and during inoculation procedures, plates should be handled in a manner that minimizes product exposure to the environment. Product which has exceeded the assigned expiration date noted on the label should not be used.

Do not use plates that exhibit evidence of drying, cracking, discoloration, microbial contamination or any other signs of deterioration. The presence of excessive condensate may indicate plates which have been damaged by exposure to temperature extremes.

VII. SPECIMEN COLLECTION

The quality of culture results depends primarily on the adequacy and condition of the specimen submitted for examination. Proper specimen collection techniques must be followed to ensure the most accurate culture results. Sterile swabs and collection containers should be used. Plates should be inoculated promptly after specimen collection. If a delay in inoculation is unavoidable, transport medium should be employed. Specimens should be collected prior to the initiation of antimicrobial therapy. Detailed information on proper specimen collection may be obtained from microbiology reference materials.[4,5]

VIII. MATERIALS PROVIDED

Modified Thayer Martin Agar Plates and lot specific Quality Control Certificate.

IX. MATERIALS REQUIRED BUT NOT PROVIDED

Incubator maintaining 35-37°C.
CO_2 environment generation (See PROCEDURE)
Ancillary culture media, reagents and laboratory equipment as required.

X. PROCEDURE

Pathogenic *Neisseria* species, especially *N. gonorrhoeae*, are fastidious organisms that exhibit sensitivity to desiccation and temperature extremes. To optimize recovery from clinical specimens, it is recommended that the media be inoculated directly with the specimen at the clinic site and immediately incubated in an atmosphere of approximately 3-10% CO_2 (atmospheric incubator or zip-lock bag with generator) at 35-37°C. Alternatively, inoculate the specimen as soon as possible after it is received in the laboratory. If material is being cultured directly from a swab, roll the swab over the entire agar medium to ensure contact with all swab surfaces. Streak over swab areas for further isolation especially if specimen contains mucous material. Avoid applying excessive pressure to the agar surface during inoculation to prevent gouging and splitting of the agar media. (Note: Agar surfaces should be smooth and moist but free of excessive moisture which could cause confluent growth patterns.)

Plates should be incubated in an atmosphere of increased CO_2 (3-10%), approximately 70% humidity, and a temper-ature of 35 - 37° C for up to 72 hours. These conditions can be met with a candle extinction jar, CO_2 generating systems, or an atmospheric incubator.

Examine plates for growth after overnight incubation, and again after 48 hours. Plates should be incubated for up to 72 hours before a negative interpretation can be made. Subculture for definitive identification of *N. gonorrhoeae* should be made within 18 - 24 hours of isolation.

XI. EXPECTED RESULTS

NCCLS CONTROL ORGANISMS (ATCC STRAINS)

Neisseria gonorrhoeae (ATCC 43069)	Growth
Neisseria meningitidis (ATCC 13090)	Growth

Proteus mirabilis (ATCC 43071)	Inhibition (partial)
Neisseria sicca (ATCC 9913)	Inhibition (complete)
Candida albicans (ATCC 60193)	Inhibition (partial)
Escherichia coli (ATCC 25922)	Inhibition (partial)

XII. LABORATORY RESULTS

Growth of *N. gonorrhoeae* should be evident after 18-72 hours of incubation. Isolates from uro-genital specimens that exhibit typical colonial morphology, a positive oxidase test, and Gram stain as a gram negative diplococcus may be presumptively identified as *Neisseria gonorrhoeae*. Specimens from other body sites have a higher incidence of isolating *N. meningitidis*.[5]

Confirmatory tests, such as rapid fermentation, carbohydrate degradation, chromogenic enzyme-substrates, etc., are recommended for all isolates from uro-genital sources and are required for all other body sites. Additional biochemical information may be obtained from reference microbiology texts.[6,7]

XIII. LIMITATIONS

The ability to detect microorganisms by culture techniques can be affected by the following factors: improper specimen collection, storage and inoculation , initiation of antiinfective therapy prior to specimen collection, improper culture incubation temperatures and atmospheres, improper length of culture incubation, and improper storage and handling of culture media.

Some strains of *N. gonorrhoeae,* inhibited by vancomycin and trimethoprim lactate, have been reported. Certain oxidase positive, gram negative bacilli will grow on selective media and produce colonies resembling *N. gonorrhoeae*.

XIV. REFERENCES

1. Martin, J.E., T.E. Billings, J.F. Hackney, and J.D. Thayer. 1967. Primary isolation of *N. gonorrhoeae* with a new commercial medium. Public Health Rep. 82:361-363.
2. Thayer, J.D. and J.E. Martin. 1966. Improved medium selective for cultivation of *N. gonorrhoeae* and *N. meningitidis* . Public Health Rep. 81:559-562.
3. Seth, A. 1970. Use of trimethoprim to prevent overgrowth by *Proteus* in the cultivation of *N. gonorrhoeae*. Brit. Journ. of Vener. Dis. 46:201-202.
4. Kellogg, D.S. Jr., K.K. Holmes, and G.A. Hill. 1976. Cumitech 4, Laboratory Diagnosis of Gonorrhea. Coordinating ed., S. Marcus, and J.C. Sherris. American Society for Microbiology, Washington, D.C.
5. Eschenbach, D., H.M. Pollock, J. Schachter. 1983. Cumitech 17, Laboratory Diagnosis of Female Genital Tract Infections. Coordinating ed., S.J. Rubin. American Society for Microbiology, Washington, D.C.
6. Koneman, E.S., S.D.Allen, V.R. Dowell, Jr. and H. M. Sommers. 1983. Color Atlas and Textbook of Microbiology, 2nd ed. J.B. Lippincot Company, Philadelphia.
7. Lennette, E.H., ed. 1985. Manual of Clinical Microbiology, 4th ed. American Society for Microbiology, Washington, D.C.

USER QUALITY ASSURANCE/ QUALITY CONTROL PROCEDURES AND INFORMATION

HealthLink recommends that the following quality assurance and quality control procedures be performed on each batch of product.

I. QUALITY ASSURANCE

The following quality assurance procedures must be performed to assure the product will perform according to its intended use within the assigned expiry date:

1. Daily document the that product storage refrigerator maintains temperature within the recommended range: 2-8°C.
2. Daily, document that laboratory incubator maintains temperature within the recommended range: 35-37°C.

II. QUALITY CONTROL

The following incoming inspection procedures must be performed for each batch (batch = same lot, same shipment) of culture media received in the laboratory :

1. Inspect plates according to instructions contained on the "Quality Control Log Sheet." (See also Section VI, "STORAGE/SHELF LIFE")
2. Peel off the lower portion of a product bag label (Quality Control Certificate) for the lot being accepted into the laboratory and affix it to the Quality Control Log Sheet.
3. Initial and date the Quality Control Log Sheet.

Note: Notify Technical Service immediately if media does not meet the inspection criteria.

This media is not exempt from end user quality control testing relating to media growth characteristics according to NCCLS Document M22A , *Quality Assurance for Commercially Prepared Microbiological Culture Media.* Quality control organisms may be ordered by contacting our customer service department.

TECHNICAL SERVICE

HealthLink provides a toll free technical service line (1-800-638-2625) to assist with product usage. To receive additional product information, procedural instructions, QA/QC log sheets, Material Safety Data Sheets, or to have technical questions answered; please call between the hours of 9:00 am to 5:00 pm EST.

HealthLink
7779 Bayberry Road
Jacksonville, FL 32256
1-800-638-2625

January, 1997

Product No. 1100 Rev. No. 00