Introduction

Intended Use

Mueller Hinton Chocolate Agar is for use in qualitative procedures for the isolation and cultivation of fastidious organisms, particularly *Haemophilus* species.

Description and Principle

Because growth of fastidious organisms is poor on Mueller Hinton basal agars, the use of Mueller Hinton Agar supplemented with 1% hemoglobin and a defined supplement, known as Mueller Hinton Chocolate Agar, has been adopted for testing of *H. influenza*.

Reagents and Appearance

InTray® Mueller Hinton Chocolate agar contains beef extract, vitamins and mineral enrichment, acid hydrolysate of casein, cystine, tryptophan, starch, agar and hemoglobin. Agar appears chocolate brown with a final pH of 7.3 ± 0.2 at 25° C.

Precautions, Safety and Disposal

For In Vitro Diagnostic Use

Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing and gloves.

Once the tray has been inoculated and resealed, re-open only in a biological safety cabinet. Because of the potential for containing infectious materials, the tray must be destroyed by autoclaving at 121°C for 20 minutes.

Storage

Upon receipt, store InTray Mueller Hinton Chocolate Agar under refrigeration (2-8°C). Avoid freezing or prolonged storage at temperatures above 40°C. Do not use InTray Mueller Hinton Chocolate Agar if the medium shows signs of deterioration or contamination.

Shelf Life

Expiration is 12 months past the date of manufacture.

Procedure

Materials Provided

InTray Mueller Hinton Chocolate Agar

1 Prepare InTray



Pull back the lower right corner adjacent to the clear window of the InTray label until the protective seal is completely visible.

Remove the seal by pulling the tab. Discard the seal.

DO NOT REMOVE OR ALTER THE WHITE FILTER STRIP OVER THE VENT HOLE!

Materials Required but Not Provided

- Sterile inoculating tool (e.g., cotton swab/forceps/scalpel blade)
- Laboratory incubator capable of incubation at 35 ± 2°C
- 5-10% CO₂ for fastidious organisms.

2 Open Seals



Inoculate the specimen on the surface of the medium. A sterile inoculating loop that has been moistened by touching the surface of the medium may be used for inoculation of solids or scrapings.

Reseal the InTray by pressing together the edges of the label against the plastic tray. Press all around the InTray to insure a complete seal. Immediately label the InTray with patient or sample information and date.

DO NOT COVER THE VIEWING WINDOW.

Incubation

Incubate the InTray according to procedures outlined in the testing standards being performed. Generally cultures are incubated at $35\pm2\,^{\circ}\text{C}$ for 18-72 hours in ambient air or in an atmosphere of 5-10% CO $_2$ for fastidious organisms.

Quality Control

This product has been tested and meets the CLSI (formerly NCCLS) Approved Standard for commercially prepared media (M22-A3). At the time of manufacture, quality control testing is performed on each lot of the InTray Mueller Hinton Chocolate Agar. The ability of the media to support growth and demonstrate expected biochemical reactions and morphology is verified by lot.

Testing of control organisms should be performed in accordance with established laboratory quality control procedures. The following QC strains are recommended for customers who choose to complete independent QC testing of the InTray Mueller Hinton Chocolate Agar

Organism	ATCC [®]	Expected Result	
H. influenziae	10211	Growth	
N. meningitides	13090	Growth	
S. pneumoniae	6305	Growth	

Reading the Results

Evaluation

Follow standard methods, such as those outlined in CLSI standards.¹

Limitations

InTray Mueller Hinton Chocolate Agar is an agar medium that is susceptible to condensation collection within the inner seal, especially when stored at low temperatures and/or having been exposed to extreme temperature fluctuations. If moisture is visible on the surface of the InTrays, dry them (with the seal removed and InTray label in a position allowing for air flow) under a BSL-2 cabinet just prior to inoculation. There should be no visible droplets of moisture on the surface of the agar when they are inoculated. The surface of the dried medium should be smooth and should not show signs (webbed ribbing pattern on the agar surface) of desiccation.

References

 CLSI. Performance Standards for Antimicrobial Disk Susceptibility Tests; Approved Standard—Twelfth Edition. CLSI document MO2-A12. Wayne, PA: Clinical and Laboratory Standards Institute; 2015. Symbol glossary: biomeddiagnostics.com/l/symbol-glossary

Document Revision History

Rev. C, May 2025

Removed QR Codes for the certificate and product information, updated manufactured by and company address.

Rev. D, Septemeber 2025

Removed ® replaced with TM.



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InTray Mueller Hinton Chocolate Agar



12-483-001



REF

12-483-002



Not available in all countries; please inquire. For *In Vitro* Diagnostic Use





