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Technical Data Sheet

Mueller Hinton Agar (MML-MHA-01)

Principle
Mueller Hinton agar duplicates the formula recommended by Mueller and Hinton (1941) for the primary isolation of *Neisseria species*. Commonly it is used for performing antibiotic susceptibility tests using a disc diffusion assay and due to its susceptibility for microorganism and high reproducibility; WHO Committee recommended it for standardization of susceptibility testing. Media is simple and composed of meat infusion form (equivalent of beef infusion form), Casamino acids, starch and agar. Meat Infusion from and casamino acids provides nitrogen vitamins, amino acids, minerals, carbon and other nutrients to support the growth of microorganisms. While starch absorb any toxic metabolites produced during microbial growth and while autoclaving undergoes hydrolysis and liberate small amount of dextrose, which act as source of energy. Agar is the solidifying agent.

Use: Recommended for antimicrobial susceptibility testing of rapidly growing aerobic and facultative anaerobic microbes by the disc diffusion assay.

Contents*

Ingredients	**Gram/Litre**
Meat Infusion Form#	300.000
Casamino Acids	17.500
Starch	1.500
Agar	17.000
pH at 25˚C	7.3 ±0.2

* Formula adjusted for optimum performance and parameters
# Equivalent to Beef infusion form

Directions: Dissolve 38.00 grams in 1000 ml distilled water. Boil to dissolve the medium completely and sterilize by autoclaving at 15 lbs. pressure (121 °C) for 15 min, cool it to 42-45 °C and distribute aseptically in petri plates. Ensure complete solidification and inoculate test sample aseptically.

Specimens types analyzed
Pharmaceutical samples, clinical and non-clinical samples etc.

Precautions to be taken
These microbial media are intended for the in-vitro use only. All the handling, experiments, storage, and discarding should be performed with the help of skilled and knowledgeable technicians and as per the established guidelines. The material should be disposed only after proper sterilization by autoclaving. Please go through the MSDS of the media to avoid any accidents or in emergency.

Performance and Evaluation
The expected performance of the medium is liable to use as per the direction on the label when stored at optimum conditions and within expiry date.

Quality Control

Appearance	Beige colored free flowing, homogeneous powder
Reaction of 3.8% solution	7.3 ±0.2 at 25 ˚C

pH	7.10- 7.50
Gelling	Firm comparable with 1.7% agar gel
Color and clarity of ready medium	Light amber opalescent gel
Growth Promotion properties	Best at ≤ 100 CFU at 32-37 ºC for 18-72 h
Indicative properties	Optimum at ≤ 100 CFU at 32-37 ºC for 18-48 h
Negative control	Performed using sterile distilled water

Different Microbial Response

Recovery is compared with Sabouraud Dextrose Agar

Organism	ATCC	Inoculum	Growth	Recovery	Incubation Temperature	Incubation period
Escherichia coli	8739	50-100	Luxurious	80-90%	33-37 ℃	18-48 h
Staphylococcus aureus	25923	50-100	Luxurious	80-90%	33-37 ℃	18-48 h
Pseudomonas aeruginosa	27853	50-100	Luxurious	80-90%	33-37 ℃	18-48 h

Antibiotic Sensitivity test

Ciprofloxacin 5mcg discs were tested for standard ATCC strains and zone of inhibition were measured after an incubation 30-35°C for 18 hours.

Organisms	Standard zone of inhibition	Observed zone of inhibition	Growth
Staphylococcus aureus (ATCC 25923)	22-30 mm	22-30 mm	Luxurious
Pseudomonas aeruginosa (ATCC 27853)	30-40 mm	30-40 mm	Luxurious

Storage and Shelf Life

Hygroscopic; keep container tightly closed. Store in cool dry place.

Disposal: To avoid the contamination or propagation of any hazardous microbes the used, unusable or modified preparation of this product must be disposed after autoclaving after completion of task.

Reference

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3. *Difco Manual* (1998). 11th Edition. Difco Laboratories., Division of Becton Dickinson and Company, Sparks, Maryland, USA.
4. Mueller, J. H., and J. Hinton (1941). *A protein-free medium for primary isolation of gonococcus and meningococcus.* Proc. Soc. Exp. Biol. Med. 48:330-333.
5. Rand, M. C., Arnold E. Greenberg, and Michael J. Taras, (1976), *Standard methods for the examination of water and wastewater*. Prepared and published jointly by American Public Health Association, American Water Works Association, and Water Pollution Control Federation.
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