Mueller Hinton Agar II

LAB 39

Description
A medium for antimicrobial sensitivity testing by the disc diffusion method. This medium, used in the technique of Bauer and Kirby, has been adopted by the National Committee for Clinical Laboratory Standards (NCCLS) in the USA as the definitive method for susceptibility testing. The medium has a very low thymine and thymidine content, making it suitable for trimethoprim and sulphonamide testing, controlled to ensure correct zone sizes with aminoglycoside and tetracyline antibiotics. The medium was originally formulated as a heat labile protein free medium for the isolation of pathogenic *Neisseriaceae.*

Formula	g/litre
Beef Extract	2.0
Acid Hydrolysed Casein	17.5
Starch	1.5
Agar No. 1	17.0
Calcium ions	50-100mg/litre
Magnesium ions	20-35mg/litre

Method for reconstitution
Weigh 38 grams of powder, disperse in 1 litre of deionised water. Allow to soak for 10 minutes, swirl to mix then sterilise at 121°C for 15 minutes. Cool to 47°C, mix well and pour plates.

Appearance: Straw coloured, clear gel.

pH: 7.3 ± 0.1

Minimum Q.C. organisms: *E. coli* ATCC 25922
S. aureus (antibiotic sensitivity zones) ATCC 25923

Storage of Prepared Medium: Plates – up to 7 days at 2-8°C in the dark.

Inoculation: Surface, inoculum as described by N.C.C.L.S.

Incubation: As recommended by methodology for particular organisms and antibiotics by NCCLS.

References
Mueller, J.H. and Hinton, J. (1941). Protein-free medium for primary isolation of gonococcus and meningococcus. Proc. Soc. Exp. Biol. and Med., 48: 330-333.

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American Public Health Association. (1950). Diagnostic Procedures and Reagents. 3rd edn., A.P.H.A., New York.

NCCLS. (1986). Performance standards for antimicrobial susceptibility testing – second informational supplement.