Middlebrook 7H9 Broth

Medium used for the cultivation and enumeration of Mycobacteria.

• CONTENTS (Liter)

Ammonium Sulfate	0.5 g
Disodium Phosphate	2.5 g
Monopotassium Phosphate	1.0 g
Sodium Citrate	0.1 g
Magnesium Sulfate	0.05 g
Calcium Chloride	0.0005 g
Zinc Sulfate	0.001 g
Copper Sulfate	0.001 g
Ferric Ammonium Citrate	0.04 g
L-Glutamic Acid	0.5 g
Pyridoxine	0.001 g
Biotin	0.0005 g

Final pH = 6.6 ± 0.2 at 25°C

• PROCEDURE

Suspend 4.7 G of powder in 900 mL of distilled or deionized water. Add 2 mL of Glycerol supplement (MB-G1821) or 0.5 mL of Tween 80 supplement (MB-T1861). Heat to boiling until completely dissolved. Sterilize by autoclave at 121°C for 10 minutes. Cool to 45 - 50°C in water bath. Aseptically add 2 vials of Middlebrook ADC Enrichment supplement (MB-M3017). Mix well. Pour into tubes.

Middlebrook ADC Enrichment supplement

1 vial contents (each vial is sufficient for 500 mL of medium)

Bovine Albumin Fraction V	2.5 g
Dextrose	1.0 g
Catalase	0.0015 g

• INTERPRETATION

Middlebrook 7H9 Broth is a medium used for the cultivation and enumeration of Mycobacteria. Ammonium sulfate provides the nitrogen source. Phosphates are the buffering agents. Sodium citrate maintains inorganic cations in the medium. Magnesium sulfate, calcium chloride, zinc sulfate, copper sulfate and ferric ammonium citrate are inorganic salts essential for the growth of Mycobacteria. L-glutamic acid, pyridoxine and biotin, glycerol and tween 80 contribute to the growth of Mycobacteria. Bovine albumin acts as the protective agent. Dextrose is the carbon and energy source. Catalase removes toxic peroxides. Growth of Mycobacteria is stimulated by 5 - 10% of CO_2 in this medium.

• TECHNIC

Inoculate the specimen using a sterile needle to the medium. Incubate at 36 ± 1°C for 2 - 4 weeks up to 8 weeks under microaerobic condition. Slightly loosen the caps. Refer appropriate references for recommended test procedure.

• QUALITY CONTROL FOR USE

Dehydrated medium
Appearance: free-flowing, homogeneous
Color: Off white
Prepared medium
Appearance: clear
Color: light amber
Incubation conditions: 36 ± 1°C / 2 - 4 weeks up to 8 weeks under microaerobic condition / loosen the caps

Microorganism	ATCC	Growth
Mycobacterium smegmatis	607	good

• STORE

The powder is very hygroscopic. Store the powder at room temperature, in a dry environment, in its original container tightly closed and use it before the expiry date on the label. Store prepared medium at 2 - 8°C.

• REFERENCES

1. Middlebrook G. and Cohn M. L., 1958, Am. J. Public Health, 48:844
2. Middlebrook, Cohn and Schaefer. 1954. Am. Rev. Tuberc. 70:852
3. MacFaddin. 1985. Media for isolation-cultivation-identification-maintenance of medical bacteria, vol. 1. Willia ms & Wilkins, Baltimore, Md.

• PACKAGE

Cat. No : MB-M0949 Middlebrook 7H9 Broth	500 G