

Medium used for the isolation and cultivation of Mycobacteria.

# • CONTENTS (Liter)

Denerostia Direct of Coopin	10 -
Pancreatic Digest of Casein	1.0 g
Disodium Phosphate	1.5 g
Monopotassium Phosphate	1.5 g
Ammonium Sulfate	0.5 g
L-Glutamic Acid	0.5 g
Sodium Citrate	0.4 g
Ferric Ammonium Citrate	0.04 g
Magnesium Sulfate	0.05 g
Copper Sulfate	0.001 g
Pyridoxine	0.001 g
Zinc Sulfate	0.001 g
Biotin	0.0005 g
Calcium Chloride	0.0005 g
Malachite Green	0.00025 g
Agar	15.0 g
Final pH = 6.6 $\pm$ 0.2 at 25°C	

# PROCEDURE

Suspend 20.49 G of powder in 900 mL of distilled or deionized water. Add 5 mL of Glycerol supplement (MB-G1821). Heat to boiling until completely dissolved. Sterilize by autoclave at 121°C for 15 minutes. Cool to 45 - 50°C in water bath. Aseptically add 2 vials of Middlebrook OADC Enrichment supplement (MB-M3021). Mix well. Pour into petri dishes.

#### Middlebrook OADC Enrichment supplement

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

Oleic Acid	0.025 mL
Albumin Fraction V, Bovine	2.5 g
Glucose	1.0 g
Catalase	0.002 g
Sodium Chloride	0.425 g

# INTERPRETATION

Middlebrook 7H11 Agar is a medium used for the isolation and cultivation of Mycobacteria. Pancreatic digest of casein provide nitrogen, carbon, amino acids, vitamins and minerals. Disodium phosphate, monopotassium phosphate, ammonium sulfate, L-glutamic acid, ferric amonium citrate, magnesium sulfate, copper sulfate, pyridoxine hydrochloride, zinc sulfate, biotin and calcium chloride are inorganic salts essential for the growth of Mycobacteria. Sodium citrate is converted to citric acid which holds inorganic cations in solution. Malachite green serves as the selective agent to inhibit bacteria except Mycobacteria. Agar is the solidifying agent. Glycerol is the carbon and energy source. Oleic acid is necessary in the metabolism of Mycobacteria. Albumin protects Mycobacteria against toxic agents. Glucose is the carbohydrate. Catalase destroys toxic peroxides that may be present in the medium. Sodium chloride maintains the osmotic balance.

#### TECHNIC

Inoculate the specimen using a sterile loop to the medium. Incubate at  $35 \pm 2^{\circ}C$  for 2 - 5 days up to 3 weeks under microaerobic condition. Refer appropriate references for recommended test procedure.

# • QUALITY CONTROL FOR USE

<u>Dehydrated medium</u> Appearance: free-flowing, homogeneous Color: light beige with greenish tint <u>Prepared medium</u> Appearance: clear to slightly opalescent Color: greenish light amber Incubation conditions:  $35 \pm 2^{\circ}C/2 - 5$  days up to 3 weeks under microaerobic condition

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., M.L. Cohn, W.E. Dye, Russell, Jr., and D. Levy, 1960. Microbiologic procedures of value in tuberculosis. Acta. Tuberc. Scand. 38:66-81.
- 2. Dubos R. J. and Middlebrook G., 1947, Am. Rev. Tuberc., 56:334.

#### PACKAGE

Cat. No : MB-M0980 Middlebrook 7H11 Agar

500 G

