

TM 100 – DUBOS OLEIC AGAR BASE

INTENDED USE

For cultivation of *Mycobacterium tuberculosis*.

PRODUCT SUMMARY AND EXPLANATION

Mycobacterium tuberculosis, the causative agent of tuberculosis in man, is carried in airborne particles known as droplet nuclei that are generated when patients with pulmonary tuberculosis cough. Infections occur when a susceptible person inhales the droplet nuclei containing the bacterium. Mycobacteria are generally isolated on medium containing either coagulated egg as base or on media containing agar. Middlebrook and Dubos media contain agar whereas Lowenstein media contain egg. The advantage of using agar is that accompanying contaminating proteolytic organisms does not liquefy the medium. Agar medium are generally recommended for testing samples obtained from non-sterile sites. Agar containing media can be made selective by the addition of antibiotics since the media are solidified by addition of agar and not by inspissation as against egg containing media. Dubos and Middlebrook recommended Dubos Oleic Broth Base for the primary isolation and subsequent cultivation of the tubercle bacilli. On comparative studies of various media, Dubos Oleic Agar Base was found to be superior to other media for the primary isolation of the bacterium.

COMPOSITION

Ingredients	Gms / Ltr
Tryptone	0.500
L-Asparagine	1.000
Monopotassium phosphate	1.000
Disodium phosphate	2.500
Ferric ammonium citrate	0.050
Magnesium sulphate	0.010
Calcium chloride	0.0005
Zinc sulphate	0.0001
Copper sulphate	0.0001

PRINCIPLE

The medium consists of tryptone and L-asparagine as sources of nitrogen. The phosphates (together with calcium chloride) buffer the media as well as serve as sources of phosphates. Magnesium sulphate, zinc sulphate, copper sulphate and ferric ammonium citrate provide trace metals and sulphates. Dubos Oleic Agar is prepared without glycerol or dextrose to avoid growth of commensals.

INSTRUCTION FOR USE

- Dissolve 4 grams in 180 ml purified/distilled water.
- Heat to boiling to dissolve the medium completely.
- Sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes.
- Cool to 45-50°C and aseptically add 20ml of sterile Oleic Albumin Supplement and 5000 to 10000 units of Penicillin to sterile, cooled 180 ml medium.
- Mix well and dispense in sterile tubes or plates.

QUALITY CONTROL SPECIFICATIONS



Appearance of Powder : Light yellow to brownish yellow homogeneous free flowing powder.
Appearance of prepared medium : Light amber coloured, clear to slightly opalescent gel forms in Petri plates.
pH (at 25°C) : 6.6 ± 0.2

INTERPRETATION

Cultural characteristics observed in presence of 5-10% CO₂, with added sterile Oleic Albumin Supplement and 5,000-10,000 units of Penicillin.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Colony Morphology	Incubation Temperature	Incubation Period
<i>Mycobacterium avium</i>	25291	50-100	Luxuriant	>=70%	Smooth, thin, non-pigmented colonies	35-37°C	2-6 Weeks
<i>Mycobacterium gordonae</i>	14470	50-100	Luxuriant	>=70%	Smooth, yellow to orange colonies which are occasionally rough	35-37°C	2-6 Weeks
<i>Mycobacterium kansasii</i>	12478	50-100	Luxuriant	>=70%	Photochromogenic with flat, smooth/somewhat granular surface slightly undulating margins	35-37°C	2-6 Weeks
<i>Mycobacterium smegmatis</i>	14468	50-100	Luxuriant	>=70%	Rough or smooth, white dome shaped colonies	35-37°C	2-6 Weeks
<i>Mycobacterium tuberculosis H37RV</i>	25618	50-100	Luxuriant	>=70%	Flat, rough, dry and usually non-pigmented	35-37°C	2-6 Weeks

PACKAGING:

In pack size of 100 gm and 500 gm bottles.

STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

DISPOSAL













After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

REFERENCES

1. Byham, 1950, Am. J. Clin. Pathol., 20:678
2. Dubos R. J., and Middlebrook G., 1947, Am. Rev. Tuberc., 56:334



3. Forbes B. A., Sahn A. S., and Weissfeld D. F., Bailey & Scotts Diagnostic Microbiology, 10th Ed., 1998, Mosby, Inc., St. Louis, Mo.
4. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
5. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock, D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
6. Kent and Kubica, 1985, Public Health Mycobacteriology : A Guide For the Level III Laboratory, USDHHS, Center for Disease Control, Atlanta.
7. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Tenover F. C., (Ed.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.
8. Roberts A. H., Wallace R. J. and Erlich P., 1950, Am. Rev. Tuberc., 61:563.

 GMP Good Manufacturing Practices Certified	 IVD For In Vitro Diagnostic Use	 QTY. Quantity	 LOT/ B. NO. Lot / Batch Number	 REF Catalogue Number	 Manufacturer
 Temperature Unit	 EC REP Authorized Representative <small>MedNet GmbH Birkstrasse 10 48163 Muenster, Germany</small>	 European Conformity	 QR Code	 Consults Instructions for Use	 Best Before

NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.

***For Lab Use Only**
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