MIO Medium (Motility Indole Ornithine Medium)

M378

Intended Use:

Recommended for the identification of *Enterobacteriaceae* on the basis of motility, ornithine decarboxylase activity and indole production.

Composition**

Ingredients	g/ L
Peptone	10.000
Tryptone	10.000
Yeast extract	3.000
L-Ornithine hydrochloride	5.000
Dextrose (Glucose)	1.000
Bromocresol purple	0.020
Agar	2.000
Final pH (at 25°C)	6.5±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 31.02 grams in 1000 ml purified/distilled water. Heat to boiling to dissolve the medium completely. Dispense into test tubes in 5 ml amounts. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool the tubes in an upright position.

Principle And Interpretation

Motility, indole production and ornithine decarboxylation are routine biochemical tests employed during identification of *Enterobacteriaceae*. Motility can be demonstrated microscopically (hanging drop) or macroscopically (tube method), where motility is observed as a diffused zone of growth flaring out from the line of inoculation. Indole test is carried out to determine the ability of an organism to split indole from tryptophan by the tryptophanase enzyme. On reaction with Kovacs reagent, indole combines with the colour in the alcohol layer, which is visualized as a red ring (in the alcohol layer) (1). If the test organisms possess the specific decarboxylase enzyme, then ornithine is decarboxylated to putrescine, an amine, resulting in a subsequent rise in the pH of the medium towards alkalinity. This causes the pH indicator bromocresol purple to change from purple to yellow colour. MIO (Motility Indole Ornithine Medium) is used for identification of *Enterobacteriaceae* on the basis of motility, indole production and ornithine decarboxylation in a single tube. This medium was formulated by Ederer and Clark (2) and evaluated by Oberhofer and Hajkowski (3). Peptone and tryptone provide amino acids and other nitrogenous substances. Yeast extract is the source of vitamin B complex. Dextrose is the fermentable carbohydrate. Test cultures are stab-inoculated into the medium butts. Motility and ornithine decarboxylation reactions are read before testing indole production. On addition of the Kovacs reagent, colour of the medium changes to yellow. Therefore positive ornithine decarboxylase test (purple) could be misinterpreted as negative (yellow). Organisms ferment dextrose to form acid, which causes the pH indicator bromocresol purple to change from purple to yellow. Organisms possessing ornithine decarboxylase enzyme, decarboxylate ornithine to putrescine which increases the pH making it alkaline, indicated by a colour change from yellow to purple throughout the medium. Decarboxylase negative reaction is indicated by yellow colour or yellow with a purple band near the top of the medium. Indole is produced from tryptophan present in tryptone (3,4). The indole produced combines with the aldehyde present in the Kovacs reagent to form a red complex.

Type of specimen

Isolated Microorganism from clinical and non-clinical samples.

Specimen Collection and Handling:

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (5,6). After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

In Vitro diagnostic use. For professional use only. Read the label before opening the container. Wear protective gloves/ protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling clinical specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations:

1. Due to nutritional variations, some strains may show poor growth.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Light yellow to pale green homogeneous free flowing powder

Gelling

Semisolid, comparable with 0.2% Agar gel.

Colour and Clarity of prepared medium

Purple coloured clear to slightly opalescent gel forms in tubes as butts

Reaction

Reaction of 3.1% w/v aqueous solution at 25°C. pH : 6.5±0.2

pH

6.30-6.70

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 40-48 hours.

Organism	Growth	Motility	Indole production	Ornithine Decarboxylation
Escherichia coli ATCC 25922 (00013*)	luxuriant	positive, growth away from stabline causing turbidity	positive reaction, red ring at the interface of the medium	positive reaction, purple colour
# *Klebsiella aerogenes* ATCC 13048 (00175*)	luxuriant	positive, growth away from stabline causing turbidity	negative reaction	positive reaction, purple colour
Klebsiella pneumoniae ATCC 13883 (00097*)	luxuriant	negative, growth along the stabline, surrounding medium remains clear	negative reaction	negative reaction
Proteus mirabilis ATCC 25933	luxuriant	motility is temperature dependent, it is more pronounced at 20°C and almost absent at 35°C	negative reaction	positive reaction, purple colour

Key : (*) Corresponding WDCM numbers. (#) Formerly known as *Enterobacter aerogenes*

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (5,6).

Reference

1. MacFaddin J. F., 2000, Biochemical tests for Identification of Medical Bacteria, 3rd Ed., Lippincott, Williams and Wilkins, Baltimore.
2. Ederer G. M. and Clark M., 1970, Appl. Microbiol., 20:849.
3. Oberhofer J. R. and Hajkowski R., 1970, Am. J. Clin. Pathol., 54:726.
4. Ewing W. H., 1986, Edwards and Ewings Identification of *Enterobacteriaceae,* 4th Ed., Elsevier Science Publishing Co., Inc., New York.
5. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
6. 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.

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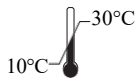
IVD ***In vitro* diagnostic medical device**



Storage temperature

EC REP CEpartner4U, Esdoornlaan 13,
3951DB Maarn, NL
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CE Marking

Do not use if package is damaged

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