

MIO Medium (Motility Indole Ornithine Medium)

Designed for the identification of Enterobacteriaceae on the basis of motility, ornithine decarboxylase activity and indole production

REF: BS.1/NZ01.100.0100	100 Gram	REF: BS.1/NZ01.250.0250	250 Gram
REF: BS.1/NZ01.500.0500	500 Gram		

PRINCIPLE AND INTERPERTATION :

Motility, indole production and ornithine decarboxylation are routine biochemical tests employed during identification of Enterobacteriaceae. Motility can be determined 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.

MEDIA COMPOSITION

Item	Formula per liter of medium
Tryptone	10.00 gm
peptone	10.00 gm
Yeast extract	3.00 gm
Dextrose	1.00 gm
L-Ornithine hydrochloride	5.00 gm
Bromocresol purple	0.02 gm
Agar	2.00 gm

Final pH 6.5 ± 0.2

** Formula adjusted, standardized to suit performance parameters.

PRECAUTIONS AND WARNINGS

Media to be handled by entitled and professionally educated person. Do not ingest or inhale.

Good Laboratories practices using appropriate precautions should be followed in:

- Wearing personnel protective equipment (overall, gloves, glasses,).
 - Do not pipette by mouth.
 - In case of contact with eyes or skin; rinse immediately with plenty of soap and water. In case of severe injuries; seek medical advice immediately.
 - Respect country requirement for waste disposal.
- S56:** dispose of this material and its container at hazardous or special waste collection point.
- S57:** use appropriate container to avoid environmental contamination.
- S61:** avoid release in environment.

For further information, refer to the MIO Medium material safety data sheet.

STORAGE AND STABILITY

BioScien MIO Medium Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°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.

PREPARATION

Suspend 31.02 g of the medium in 1 L of purified/distilled water. Heat to boiling until completely dissolved. Dispense 5 mL into test tubes and autoclave at 121°C (15 psi) for 15 minutes. Cool the tubes in an upright position before use.

Deterioration

The color of **BioScien** MIO Medium Is Light yellow to pale green homogeneous free flowing powder. prepared medium is purple coloured clear to slightly opalescent gel forms in tubes as butts

EQUIPMENT REQUIRED NOT PROVIDED

- Sterile petri dishes
- Incubator
- Autoclave

PERFORMANCE CHARACTERISTICS

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

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

QUALITY CONTROL

To ensure adequate quality control, it is recommended that positive and negative control included in each run. If control still out of range please contact **BioScien** technical support.

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.

SYMBOLS IN PRODUCT LABELLING

	For in-vitro diagnostic use		Number of <n> test in the pack
	Batch Code/Lot number		Caution
	Catalogue Number		Do not use if package is damaged
	Temperature Limitation		Consult Instruction for use
	Expiration Date		
	Manufactured by		