TM 186 - MIO MEDIUM (MOTILITY INDOLE ORNITHINE MEDIUM)

INTENDED USE

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

PRODUCT SUMMARY AND EXPLANATION

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). 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 and evaluated by Oberhofer and Hajkowski.

COMPOSITION

Ingredients	Gms / Ltr
Casein enzymic hydrolysate	10.000
Peptic digest of animal tissue	10.000
Yeast extract	3.000
L-Ornithine hydrochloride	5.000
Dextrose	1.000
Bromocresol purple	0.020
Agar	2.000

PRINCIPLE

Casein enzymic hydrolysate and peptic digest of animal tissue 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 casein enzymic hydrolysate. The indole produced combines with the aldehyde present in the Kovacs reagent to form a red complex.

INSTRUCTION FOR USE

- Dissolve 31.02 grams in 1000 ml distilled water.
- Heat to boiling to dissolve the medium completely.
- Dispense in test tubes in 5 ml amounts.

- Sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes.
- Cool the tubes in an upright position.

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder : Light yellow to pale green homogeneous free flowing powder.

Appearance of prepared medium : Purple coloured clear to slightly opalescent gel forms in tubes as butts.

pH (at 25°C) : 6.5±0.2

INTERPRETATION

Cultural characteristics observed after an incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Motility	Indole production	Ornithine Decarboxylation	Incubation Temperature	Incubation Period
Escherichia coli	25922	50-100	Luxuriant	Positive, growth away from stabline causing turbidity	Positive reaction, red ring at the interface of the medium	Positive reaction, purple colour	35-37°C	40-48 Hours
Enterobacter aerogenes	13048	50-100	Luxuriant	Positive, growth away from stabline causing turbidity	Negative reaction	Positive reaction, purple colour	35-37°C	40-48 Hours
Klebsiella pneumoniae	13883	50-100	Luxuriant	Negative, growth along the stabline, surrounding medium remains clear	Negative reaction	Negative reaction	35-37°C	40-48 Hours
Proteus mirabilis	25933	50-100	Luxuriant	Motility is temperature dependent, it is more pronounced at 20°C and Almost absent at 35°C	Negative reaction	Positive reaction, purple colour	35-37°C	40-48 Hours

PACKAGING:

In pack size of 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.

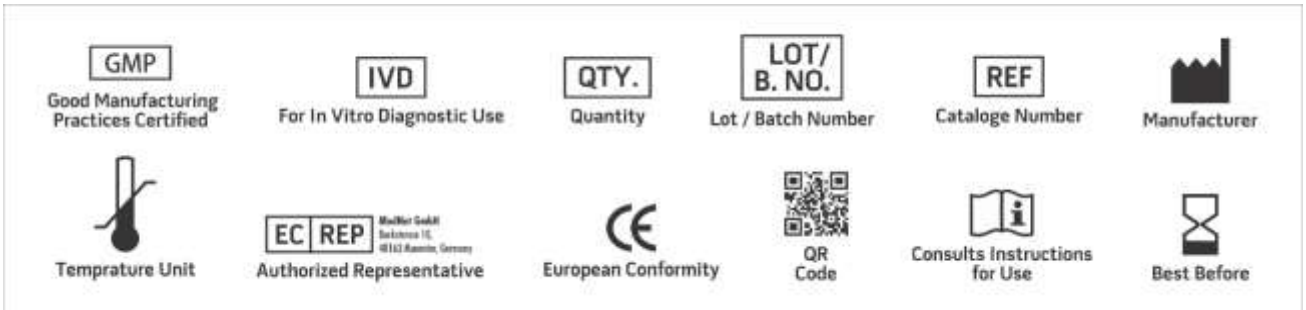
Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

DISPOSAL

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

REFERENCES

1. 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. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore.
5. Ewing W. H., 1986, Edwards and Ewings Identification of Enterobacteriaceae, 4th Ed., Elsevier Science Publishing Co., Inc., New York.



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

***For Lab Use Only**

Revision: 08 Nov., 2019