MIO Medium (Motility-Indole-Ornithine)

Cat. 1510

For the differentiation of Enterobacteriaceae

Practical information

Aplications	Categories
Differentiation	Enterobacteria

Industry: General cultivation

Principles and uses

MIO Medium (Motility-Indole-Ornithine) is a semisolid medium used for the differentiation of the Enterobacteriaceae group by motility, ornithine decarboxylase activity and indole production.

Gelatin and Casein peptones provide nitrogen, vitamins, minerals and amino acids essential for growth. They also provide tryptophan, needed for the creation of indole. Yeast extract is a source of vitamins, particularly of the B-group; Dextrose is the fermentable carbohydrate providing carbon and energy. L-ornithine is added to test the presence of the enzyme ornithine decarboxylase. If the organisms possesses such enzyme, it will be activated in an acid environment created by the initial fermentation of dextrose. Once the amino acid is decarboxylated, diamine putescine is produced. The result is an alkalinization of the medium, which turns it a dark blue. Organisms without the the enzyme, will remain acidic due to the fermentation, resulting in a yellow color in the medium. Bromocresol purple is a pH indicator to indicate decarboxylase activity; the low concentration of Bacteriological agar is for motility.

Formula in g/L

Bacteriological agar	2	Bromocresol purple	0,02
Casein peptone	10	Dextrose	1
Gelatin peptone	10	Yeast extract	3
L-Ornithine	5		

Preparation

Suspend 31 grams of the medium in one liter of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Dispense in screw-capped tubes and sterilize in autoclave at 121°C for 15 minutes

Instructions for use

Inoculate by stabbing the MIO medium and incubate in an aerobic atmosphere for 18 - 24 hours at 35 ± 2°C.

If the indole reaction is negative, incubate for an additional 24 hours. Read the motility and ornithine decarboxylase reactions before adding the Kovac's Reagent (Cat. 5205) for the indole test. The motility is indicated by cloudiness in the media or growth extending away from the line of inoculation. Ornithine decarboxylation is indicated by a purple color in the medium. A negative ornithine reaction produces a yellow color at the bottom of the tube.

*For the indole test:

- Add 3 to 4 drops of Kovac's Reagent (Cat. 5205), and shake the tube gently.
- The appearance of a red or pink color in the reagent layer is a positive indication of indole.
- Kovacs reagent detects the microorganisms capable of cleaving the tryptophan. When these microorganisms are present in the medium they liberate indole that reacts with 4-dimethylaminobenzaldehyde to form a dark red dye.
- Compare the results with a non-inoculated test tube.

Quality control

Solubility	Appareance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25ºC)
w/o rests	Fine powder	Beige	Purple	6,5 ± 0,2

Microbiological test

Incubation conditions: (35±2 ºC / 18-24 h)

Microrganisms	Specification	Characteristic reaction
Enterobacter aerogenes ATCC 13048	Good growth	Motility (+), Indole (-), Ornithine Decarboxylation (+)
Klebsiella pneumonieae ATCC 13883	Good growth	Motility (-), Indole (-), Ornithine Decarboxylation (-)
Escherichia coli ATCC 25922	Good growth	Motility (+), Indole (+), Ornithine Decarboxylation (+)
Proteus mirabilis ATCC 25933	Good growth	Motility (+), Indole (-), Ornithine Decarboxylation (+)

Storage

Temp. Min.:2 ºC
Temp. Max.:25 ºC

Bibliography

Ederer, G.M., and M. Clark. 1970. Motility-Indole-Ornithine medium. Appl. Microbiol. 2:849.
Oberhofer, T.R., and R. Hajkowski. 1970. Evaluation of non-lactose-fermenting members of the Klebsiella-Enterobacter-Serratia Division. I. Biochemical characteristics. Am. J. Clin. Pathol. 54:720.