MOTILITY-INDOLE-LYSINE MEDIUM (MIL MEDIUM) TM 225

INTENDED USE

For identification of members of Enterobacteriaceae on the basis of motility, lysine decarboxylase, lysine deaminase and indole production

COMPOSITION

Ingredients	Gms/Ltr.
Peptic digest of animal tissue	10.000
Casein enzymatic hydrolysate	10.000
L-Lysine hydrochloride	10.000
Yeast extract	3.000
Agar	2.000
Dextrose	1.000
Ferric ammonium citrate	0.500
Bromocresol purple	0.020

PRODUCT SUMMARY AND EXPLANATION

MIL Medium is prepared as per the formulation of Reller and Merrett. It is a highly useful medium in the identification of Enterobacteriaceae as it provides four differential reactions in a single culture tube. It is recommended to be used along with Triple Sugar Iron Agar and Urea Agar so as to enable presumptive identification of members of Enterobacteriaceae from faecal specimens.

PRINCIPLE

Peptic digest of animal tissue, casein enzymatic hydrolysate and yeast extract supply amino acids and other complex nitrogenous substances. A small amount of agar is added for demonstration of motility along the stab line of inoculation. Growth of motile organisms extends out from the line of inoculation, while non-motile organisms grow only along the stab line. Bromocresol purple serves as the pH indicator. Enteric gram negative bacilli ferment dextrose producing acid which changes the pH indicator to yellow. Lysine is decarboxylated by some enteric gram negative bacilli, forming the end product cadavarine. This produces a purple butt in the tube (reversal of pH). Lysine deamination produces α-ketogluteric acid and results in deep red rim at the surface of the medium and a yellow butt in the tube. Indole is produced by organisms that possess the enzyme tryptophanase which degrades tryptophan in the medium. The reaction is detected by adding Kovac's reagent to the surface of the medium. Indole combines with p-dimethylaminobenzaldehyde (Kovac's reagent) to produce red complex.

INSTRUCTION FOR USE

1. Dissolve 36.52 grams in 1000 ml distilled water.
2. Gently heat to boiling to dissolve the medium completely.
3. Dispense into tubes in 5 ml amounts.
4. Sterilize by autoclaving at 15 psi (121°C) for 15 minutes.
5. Cool the tubes in an upright position.

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder: Cream to greenish yellow colour, homogeneous free flowing powder

Appearance of prepared medium: Reddish purple colour, clear to slightly opalescent gel

pH (at 25°C): 6.6 ± 0.2

INTERPRETATION:

Culture characteristics observed after an incubation period of 18 – 24 hours at 35 ± 2°C.

Microorganisms	ATCC	Inoculum (CFU)	Motility	Indole production	Lysine Deaminase	Lysine Decarboxylation
Enterobacter aerogenes	13048	50-100	Positive, growth away from stabline	Negative	Negative	Positive reaction, purple colour
Escherichia coli	25922	50-100	Positive, growth away from stabline	Positive	Negative	Variable reaction
Proteus mirabilis	25933	50-100	Positive, growth away from stabline	Negative	Positive	Negative reaction, yellow colour
Klebsiella pneumoniae	13883	50-100	Negative, growth along the stabline	Variable	Negative	Positive reaction, purple colour
Salmonella enteritidis	13076	50-100	Positive, growth away from stabline	Negative	Negative	Positive reaction, purple colour

STORAGE & STABILITY

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers below 25°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.

REFERENCES

1. Reller L. B. and Mirrett S., 1975, J. Clin. Microbiol., 2:247.
2. Ewing W. H., 1986, Edwards and Ewings Identification of Enterobacteriaceae, 4th Ed., Elsevier Science Publishing Co., Inc., New York, N.Y.
3. Forbes B. A, Sahm A. S. and Weissfeld D. F., 1998, Bailey & Scotts Diagnostic Microbiology, 10th Ed., Mosby, Inc., St. Louis, Mo.
4. Murray P. R., Baron E. J., Jorgensen J. H., Pfaller M. A., Yolken R. H., (Eds.), 8th Ed., 2003, Manual of Clinical Microbiology, ASM, Washington, D.C.
5. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore.

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