

Mannitol-Mobility-Nitrate/Agar

355-5514
356-4874

DEFINITION

Medium used for the biochemical identification of *Enterobacteria* and coliforms.

PRINCIPLE

The principle of the medium relies on the ability of certain *Enterobacteria* to ferment mannitol (turns yellow) and to reduce nitrates to nitrites.

PRESENTATION

• Ready-to-use

10 ml x 25 tubes

code 355-5514

• Dehydrated

500 g

code 356-4874

STORAGE

- Ready-to-use : + 2°C to 8°C.
- + 15°C to 25°C, in carefully-sealed bottles in a cool, dry place.
- Expiration date and batch number are shown on the package.

THEORETICAL FORMULA

Tryptic casein hydrolysate	10 g
Potassium nitrate	1 g
Mannitol	7.5 g
Phenol red	40 mg
Agar	3.5 g
Distilled water	1,000 ml
Final pH (25°C) = 7.6 ± 0.2	

OTHER PRODUCTS REQUIRED (NOT SUPPLIED)

- Distilled water

EQUIPMENT REQUIRED (NOT SUPPLIED) (non-exhaustive)

- Hotplate
- Mixer-homogenizer
- Test tubes (16 x 160 mm) with autoclave-proof stoppers
- Water-bath, precise to + 1°C
- Thermostatically-controlled incubator or incubation room, precise to ± 1°C
- Autoclave
- All usual laboratory equipment.

PREPARATION OF DEHYDRATED MEDIUM

Always shake well before use.

Dissolve 22 g of powder in 1 liter of distilled water. Wait for 5 minutes, then mix until a homogenous suspension is obtained.

Heat gently, swirling frequently, then bring to the boil until completely dissolved. Dispense in tubes so as to obtain a unit of 6 - 7 cm.

Sterilize in autoclave at 121°C ± 1°C for 15 minutes.

Reconstitution ratio: 22 g/l

500 g of powder makes 22.7 liters of medium.

PROTOCOL

• Inoculation and incubation

As this medium is only weakly spiked with agar, if the pellet has disintegrated prior to inoculation, the medium should be melted in a boiling waterbath and left to solidify by placing the tubes vertically in cold water.

Using a platinum wire, inoculate with a central injection through to the base of the medium.

Incubate at 37°C ± 1°C for 18 - 24 hours.

READING AND INTERPRETATION

This medium yields three results:

- Mannitol fermentation: the medium turns yellow. If this is not the case, it retains its initial color.
- Mobile bacilli diffuse into the medium from the inoculation line, creating turbidity. Immobile bacilli only grow along the inoculation streak.
- By adding GRIESS reagents (sulfanilic acid and α-naphthylamine) to the surface of the medium, nitrite can be detected if the bacteria produce nitrate reductase.

N.B.: A strict nitrate-breathing aerobe (e.g. *Pseudomonas aeruginosa*, *Pseudomonas stutzeri*, *Alcaligenes denitrificans*), growing only on the surface in a meat-liver medium without nitrate, will grow throughout the mass of the Mannitol-Mobility-Nitrate medium while producing abundant gas bubbles (denitrification).

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Using the results obtained, bacteria can be identified by means of the following table:

ENTEROBACTERIACEAE	Motility	Mannitol
<i>Salmonella SE I</i> in general	+	+
<i>S. Typhi</i>	+	+
<i>S. Paratyphi A</i>	+	+
<i>S. Arizona SE III</i>	+	+
<i>Citrobacter</i>	+	+
<i>Edwardsiella</i>	+	-
<i>Escherichia coli</i>	+	+
<i>Alkalescens</i>	+	+
<i>S. dysenteria</i>	-	-
<i>S. boydii, flexneri</i>	-	+
<i>S. sonnei</i>	-	+
<i>Proteus vulgaris</i>	+	-
<i>Proteus mirabilis</i>	+	-
<i>Proteus rettgeri</i>	+	+
<i>Proteus morgani</i>	+	-
<i>Providencia</i>	+	-
<i>Levinea</i>	+	+
<i>Y. enterocolitica</i>	-	+
<i>Y. pseudotuberculosis</i>	-	+
<i>K. oxytoca</i>	+	+
<i>E. aerogenes</i>	-	+
<i>K. ozonae</i>	-	+
<i>K. rhinoscleromatis</i>	+	+
<i>E. cloacae</i>	+	+
<i>E. agglomerans</i>	+	+
<i>Hafnia alvei</i>		+
<i>Serratia marcescens</i>	+	
<i>Serratia liquefaciens</i>	+	+
<i>V. parahaemolyticus</i>		+

PRECAUTIONS

Comply with Good Laboratory Practice.

PERFORMANCES / QUALITY CONTROL OF THE TEST

The growth performances of the media are verified with the following strains:

STRAINS	Results of 18 – 24 h culture at 37°C		
	Mobility	Mannitol	Nitrate
<i>Escherichia coli</i> ATCC 25922	+	+	+
<i>Klebsiella pneumoniae</i> ATCC 13883	-	+	+
<i>Proteus mirabilis</i> ATCC 25933	+	-	+
<i>Shigella sonnei</i> ATCC 25931	-	+	+
<i>Salmonella Enteritidis</i> ATCC 13076	+	+	+

QUALITY CONTROL OF MANUFACTURER

Every product manufactured and marketed by Bio-Rad is subject to a quality-assurance procedure at all stages, from the reception of raw materials to the marketing of the end-product. Each batch of finished product undergoes quality control and is marketed only if it satisfies the acceptability criteria.

Documentation relative to the production and control of each batch is kept on file.