

Motility Test Medium

Intended Use

Motility Test Medium is used for the detection of motility of gram-negative enteric bacilli.

Summary and Explanation

In 1936, Tittsler and Sandholzer reported on the use of semisolid agar for the detection of bacterial motility.¹ Their original formulation has been modified in the medium supplied as BBL™ brand Motility Test Medium.

Principles of the Procedure

Bacterial motility can be observed directly from examination of the tubes following incubation. Growth spreads out from the line of inoculation if the organism is motile. Highly motile organisms provide growth throughout the tube. Growth of nonmotile organisms only occurs along the stab line. TTC (triphenyltetrazolium chloride) may be added to facilitate the detection of motility. TTC is a redox indicator that is colorless in the oxidized form but becomes an insoluble red precipitate when reduced.

Formula

BBL™ Motility Test Medium

Approximate Formula* Per Liter

Beef Extract.....	3.0	g
Pancreatic Digest of Casein	10.0	g
Sodium Chloride	5.0	g
Agar	4.0	g

*Adjusted and/or supplemented as required to meet performance criteria.

Directions for Preparation from Dehydrated Product

1. Suspend 22 g of the powder in 1 L of purified water. Mix thoroughly.
2. Heat with frequent agitation and boil for 1 minute to completely dissolve the powder.
3. Dispense and autoclave at 121°C for 15 minutes.
4. If desired, 5 mL of sterile 1% TTC solution may be added aseptically after autoclaving.
5. Test samples of the finished product for performance using stable, typical control cultures.

User Quality Control

Identity Specifications

BBL™ Motility Test Medium

Dehydrated Appearance:	Fine, homogeneous, free of extraneous material.
Solution:	2.2% solution, soluble in purified water upon boiling. Solution is pale to light, yellow to tan, clear to slightly hazy.
Prepared Appearance:	Pale to light, yellow to tan, clear to slightly hazy.
Reaction of 2.2% Solution at 25°C:	pH 7.3 ± 0.2

Cultural Response

BBL™ Motility Test Medium

Prepare the medium per label directions. Stab inoculate with fresh cultures and incubate at 35 ± 2°C for 2 days.

ORGANISM	ATCC™	RECOVERY	MOTILITY
<i>Enterobacter aerogenes</i>	13048	Good	+
<i>Escherichia coli</i>	25922	Good	+
<i>Klebsiella pneumoniae</i>	33495	Good	–
<i>Salmonella enterica</i> subsp. <i>enterica</i> serotype Typhimurium	14028	Good	+
<i>Shigella flexneri</i>	9199	Good	–
<i>Proteus vulgaris</i>	8427	Good	+



Procedure

Inoculate tubes with a pure culture by stabbing the center of the column of medium to greater than half the depth. Incubate tubes for 24-48 hours at $35 \pm 2^\circ\text{C}$ in an aerobic atmosphere.

Expected Results

After incubation, observe the tubes for growth in relation to the stab line. Nonmotile organisms grow only along the line of inoculation, while motile organisms spread out from the line of inoculation and may even grow throughout the medium.

Negative tubes can be reincubated at $25 \pm 2^\circ\text{C}$ for an additional 5 days, if desired.

Consult appropriate texts for results with specific organisms.^{2,3}

Limitation of the Procedure

Many organisms fail to grow deep in semisolid media; inoculating pour plates may be advantageous.⁴

References

1. Tittsler and Sandholzer. 1936. J. Bacteriol. 31:575.
2. Holt, Krieg, Sneath, Staley and Williams (ed.). 1994. Bergey's Manual™ of determinative bacteriology, 9th ed. Williams & Wilkins, Baltimore, Md.
3. Murray, Baron, Jorgensen, Landry and Pfaller (ed.). 2007. Manual of clinical microbiology, 9th ed. American Society for Microbiology, Washington, D.C.
4. MacFaddin. 1985. Media for isolation-cultivation-identification-maintenance of medical bacteria, vol. 1. Williams & Wilkins, Baltimore, Md.

Availability

BBL™ Motility Test Medium

BAM **CCAM** **COMPF** **USDA**

Cat. No.	211436	Dehydrated – 500 g
	221509	Prepared Tubes – Pkg. of 10
	221510	Prepared Tubes – Ctn. of 100

Difco™ TTC Solution 1%

Cat. No.	231121	Tube – 30 mL
	264310	Bottle – 25 g