Motility Medium S Base

M514

Motility Medium S Base is recommended for easy detection of bacterial motility by means of TTC reduction.

Composition**

Ingredients	Gms / Litre
Beef heart, infusion from	500.000
Tryptose	10.000
Gelatin	30.000
Sodium chloride	5.000
Dipotassium phosphate	2.000
Potassium nitrate	2.000
Agar	1.000
Final pH (at 25°C)	7.3±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 60 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45 - 50°C and add 10 ml of 1% solution of 2, 3, 5-Triphenyl Tetrazolium Chloride (TTC Solution 1%, FD057). Mix well and dispense in sterile tubes.

Principle And Interpretation

Bacterial motility is an important determinant in making a final species identification. Bacteria translocate by means of flagella, the number and location of which may vary among different species. Interpretation of motility test is done by macroscopic examination of the motility medium for a diffused zone of growth flaring out from the line of inoculation (1).

Motility Medium S Base is formulated as per Ball and Sellers (2), and is used to determine motility, gelatin liquefaction and nitrate reduction. All the tests can be determined using a single tube.

Beef heart infusion and tryptose provide nitrogenous compounds, sulphur, carbon and other essential growth nutrients. Sodium chloride maintains osmotic equilibrium. 0.1% agar in presence of 3% gelatin is sufficient to preserve an intact stab line. Nitrate reduction is tested using nitrate reagents after recording motility results (3). Motility is observed as diffused growth away from the stab inoculation line while non-motile organisms grow along the stab line. The use of TTC aids in the visual detection of bacterial motility. Tetrazolium salts are colourless but are converted into insoluble red formazan complexes by the reducing properties of growing bacteria. Development of this red colour helps to trace the spread of bacteria from the inoculation line. However these salts may inhibit certain fastidious bacteria and cannot be used in all cases (1). Potassium nitrate serves as a substrate for nitrate reaction. Organisms capable of reducing nitrate exhibit increased motility in presence of 0.2% potassium nitrate, especially nitrate-reducing obligate aerobes (2). Phosphate maintains buffering in the medium and it also has a stimulatory effect on motility of *Proteus* species (2). Organisms having the ability to produce gelatinase digest or liquefy gelatin. Gelatin liquefaction can be determined by placing the test medium tubes in a refrigerator or ice bath, after an incubation at 35-37°C (4).

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Gelling

Semisolid, comparable with 0.1% Agar gel and 3.0% Gelatin gel.

Colour and Clarity of prepared medium

Yellow coloured clear to slightly opalescent gel forms in tubes as butts

Reaction

Reaction of 6.0% w/v aqueous solution at 25°C. pH : 7.3±0.2

pH
7.10-7.50

Cultural Response
M514: Cultural characteristics observed with added 1% TTC solution (FD057)after an incubation at 35-37°C for 18-48 hours.

Organism	Inoculum (CFU)	Growth	Motility	TTC reduction
Enterobacter aerogenes ATCC 13048	50-100	good-luxuriant	positive, growth away from stabline causing turbidity	positive reaction, red to maroon colour
Escherichia coli ATCC 25922	50-100	good-luxuriant	positive, growth away from stabline causing turbidity	positive reaction, red to maroon colour
Klebsiella pneumoniae ATCC 13883	50-100	good-luxuriant	negative, growth along the stabline, surrounding medium remains clear.	positive reaction, red to maroon colour
Proteus mirabilis ATCC 25933	50-100	good-luxuriant	negative, motility is temperature dependent. It is more pronounced at 20°C and almost absent at 35°C	positive reaction, red to maroon colour

Storage and Shelf Life
Store below 30°C in tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label.

Reference
1. Koneman E. W., Allen S. D., Janda W. M., Schreckenberger P. C., Winn W. C. Jr., 1992, Colour Atlas and Textbook of Diagnostic Microbiology, 4th Ed., J. B. Lippinccott Company
2. Ball R. J. and Seller W., 1966, Appl. Microbiol., 14 (4): 670.
3. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. I, Williams and Wilkins, Baltimore.
4. MacFaddin J. F., 2000, Biochemical tests for Identification of Medical Bacteria, 3rd Ed., Lippincott, Williams and Wilkins, Baltimore.

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