Sellers Agar

Cat. 1065

Differential medium for studies of gram-negative, non fermenting bacteria.

Practical information

Aplications	Categories
Detection	Non fermentative gram negative bacteria
Differentiation	Non fermentative gram negative bacteria

Industry: Clinical

Principles and uses

Sellers Agar is a very useful medium to identify and differentiate gram-negative, non-fermenting bacilli, such as Pseudomonas aeruginosa and Alcaligenes faecalis. The differentiation is based on the detection of fluorescence, glucose oxidation, production of nitrogen gas and pH changes, from clinical samples and other materials.

Gelatin peptone provides nitrogen, vitamins, minerals and amino acids essential for growth. Yeast extract is a source of vitamins, particularly of the B-group. Magnesium sulfate is a cofactor for various metabolic reactions. Sodium nitrite provides nitrogen to some organisms. L-Arginine provides amino acids for growth. Sodium chloride maintains the osmotic balance. Dipotassium phosphate acts as a buffer system. D-Mannitol fermentation is detected by Bromothymol blue as a yellow halo around the colonies. Phenol red is a pH indicator. Bacteriological agar is the solidifying agent.

Acinetobacter calcoaceticus morphologically resemble Neisseria and are frequently erroneously reported as causes of gonococcal urethritis and meningococcal meningitis (resistant to penicillin). To aid in the identification of the non-fermenters, other media such as OF Basal Medium (Cat. 1500), Indole Nitrate Medium (Cat. 1504), etc., should be used.

Under UV light only the Pseudomonas exhibit fluorescence, which is stimulated by magnesium and mannitol in the medium. At times, it is necessary to hold the tubes for 2 days for Pseudomonas to produce a typical alkaline (blue color) reaction in the medium. After incubation, check for glucose oxidation by the appearance of a yellow band, which can disappear after 24 hours.

Formula in g/L

Bromthymol blue	0,04	Bacteriological agar	13,5
Dipotassium phosphate	1	D-mannitol	2
Gelatin peptone	20	L-Arginine	1
Magnesium sulfate	1,5	Phenol red	0,008
Sodium chloride	2	Yeast extract	1
Sodium Nitrate	1	Sodium nitrite	0,35

Preparation

Suspend 43,4 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 into tubes and sterilize in autoclave at 121 ºC for 10 minutes. Allow to cool in a slanted position in order to obtain butts of 3,5 cm depth and a slant length of 7-7,5 cm.

Important: Immediately before inoculation, add 0,15 ml or 2 drops of 50 % dextrose aqueous solution, allowing it to run down the side of the tube opposite to the slant.

Instructions for use

Inoculate the medium by stabbing the base of the tube with a needle and streaking the slant. Incubate at 35±2 °C for 18-24 hours.

Quality control

Solubility	Appareance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25ºC)

w/o rests	Fine powder	Veige	Green	6,7±0,2

Microbiological test

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

Microorganisms	Specification	Characteristic reaction
Acinetobacter calcoaceticus ATCC 19638	Good growth	Blue slide, Green butt, Yellow strip, Fluorescence (-)
Pseudomonas aeruginosa ATCC 27853	Good growth	Blue-green slide, Blue-green butt, Blue strip, Fluorescence (+), Gas/Nitrogen (+)
Alcaligenes faecalis ATCC 8750	Good growth	Blue-green slide, Blue-green butt, Fluorescence (-), Gas/Nitrogen (+)

Storage

Temp. Min.:2 ºC
Temp. Max.:25 ºC

Bibliography

Sellers J. Bact. 87: 46. 1964 Lennette E.H., Spaulding H.E. and Truant P.J. Manual of Clinical Microbiology, 2nd Ed. 1974.