Technical Information

Cetrimide Agar

Product Code: DM 1024H

Application:- Cetrimide Agar is a selective medium used for the isolation of pseudomonas areuginosa from pharmaceutical products in accordance with the microbial limit testing by harmonized methodology of BP.

Composition**

Ingredients	Gms / Litre
Pancreatic digest of gelatin	20.000
Magnesium chloride	1.400
Dipotassium sulphate	10.000
Cetrimide	0.300
Agar	13.600
pH after sterilization (at 25°C)	7.2±0.2

** Formula adjusted, standardized to suit parameters

Principle & Interpretation

Cetrimide Agar was detailed by king et al [1]. This media is based on the composition described in BP[3]. and is accordance With the harmonized method of USP/EP/IP/JP [2,4,5,7]. It is used as a selective medium for the isolation of pseudomonas areuginosa from pharmaceutical products. This medium is also used for microbial limit testing for non-sterile products.

Lowburry first reported the use of cetrimide as an agent for selective isolation of pseudomonas [6]. This Medium is also used for Determining the ability of an organism to products fluorescein and pyocyanin. Cetrimide (N-acetyl-N,N,N- trimethylammonium bromide) Is added in the medium to inhibit bacteria other than pseudomonas areuginosa. This is a cationic detergent and acts as a Quaternary ammonium compound, Which causes nitrogen and phosphorus to be released from bacterial cells except pseudomonas aeruginosa. Magnesium chloride and potassium sulphate present in the medium enhances the production of pigment Pyocyanin,which is a blue-green pigment, diffusing in to the medium. This improves detection of pseudomonas on this medium Presence of magnesium ions can also neutralize EDTA, if present in the sample. Pancreatic digest of gelatin provides the essential nutrients for growth of pseudomonas,While glycerin serves as slow and continuous carbon source for the growing cell.

For the isolation of pseudomonas aeruginasa, plantes of cetrimide Agar should be inoculated from non- selective medium such as soyabean casein Digest medium (DM1011H). If the count is hight the test sample can be derectly inoculated onto this medium. Pseudomonas aeruginasa colonies may appear pigmented greenish (underUV light also). Addition of nalidixic acid can aid in Inhibiting the growth of accompanying flora.

Methodology

Suspend 45.3 grams of powder media in 1000 ml purfd/distilled water containing 10 ml glycerol. Shake well & heat to dissolve the medium completely. Steritize by autoclaving at 15 lbs pressure (121°C) for 15 minutes or as per validated cycle.

Quality Control

Physical Appearance

Cream to yellow homogeneous free flowing powder

Gelling

Firm, comparable with 1.36% Agar gel

Colour and Clarity of prepared medium

Light amber coloured opalescent gel with a slight precipitate forms in petri plates

pH range

7.00-7.40

Growth Promoting Test

Growth Promotion is carried out accordance with the harmonized method of BP. Cultural response was observed after an incubatton at 30-35°C for specified time. Recovery rate is considered as 100% for bacteria growth on soyabean casein Digest Agar.

Growth promoting properties

Growth of microorganism comparable to that previously obtained with preciously tested and approved lot of medium occurs at the specicied temperature for not more than the shortest period of time specified inculating <=100cfu (at 30935°C for <=18hours).

Inhibitory propertied
NO growth of the test microorganism occurs for the specified temp for less than longest period of time specified inoculating >=100 cfu (at 30°C for <=18 hours).
Cultural Response/Characteristics
DM 1024H: Cultural characteristics observed after incubation at30-35°C for 18-72 hours. Recovery rate is considered as 100% for bacteria growth on soyabean casein Digest Agar.

Organism	Inoculum (CFU)	Growth	Observed Lot value (CFU)	Recovery	Incubation temperature	Incubation period
Pseudomonas aeruginosa ATCC 9027	50-100	luxuriant	25-100	>=50%	30-35°C	<=18hrs
Inhibitory						
Escherichia coli ATCC 8739	>=10^3	Inhibited	0	0%	30-35°C	>=72hrs
Additional Microbiological testing						
Pseudomonas areuginosa ATCC27853	50-100	luxuriant	25-100	>=50%	30-35°C	18-24hrs
Pseudomonas areuginos ATCC 25668	>=10^3	Inhibited	25-100	>=50%	30-35°C	18-24hrs
Stenotrophomonas maltophila ATCC 13637	>=10^3	Inhibited	0	0%	30-35°C	>=72hrs
Escherichia coli ATCC 25922	>=10^3	Inhibited	0	0%	30-35°C	>=72hrs
Escherichia coli NCTC 9002	>=10^3	Inhibited	0	0%	30-35°C	>=72hrs
Staphylococcus aureus ATCC 6538	>=10^3	Inhibited	0	0%	30-35°C	>=72hrs
Sammonella Typhimurium ATCC 14028	>=10^3	Inhibited	0	0%	30-35°C	>=72hrs
Proteus mirabilis ATCC 29906	>=10^3	Inhibited	0	0%	30-35°C	>=72hrs

Storage and Shelf Life

Dried Media: Store below 30°C in tightly closed container and use before expiry date as mentioned on the label.
Prepared Media: 2-8^0 in sealable plastic bags for 2-5 days.

Further Reading

1. King, Ward and Raney, 1954, J. Lab Clin. Med., 44: 301.
2. The United States Pharmacopoeia, 2011, The United States Pharmacopoeial Convention. Rockville, MD.
3.British Pharmacopoeia 2011, The Sationery Office British Pharmacopoeia
4.European Pharmacopoeia 2011, European Dept. for the quality of Medicines,
5.Japnese pharmacopoeia, 2008
6. Lowbury EJL., 1951,J, Clin Path., 4:66.
7.Indian Pharmacopoeia, 2010 Ministry of Health and family Welfare, Govt. of India

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