Clausen Medium

M552

Clausen Medium is recommended by Nordic Pharmacopoeia Board for sterility testing.

Composition**

Ingredients	Gms / Litre
Casein enzymic hydrolysate	15.000
Papaic digest of soyabean meal	3.000
Yeast extract	6.000
Dextrose	6.000
Sodium chloride	2.500
Dipotassium phosphate	2.000
Sodium citrate	1.000
L-Cystine	0.500
L-Asparagine	1.250
Sodium dithionite	0.400
Sodium thioglycollate	0.500
Lecithin	0.300
Magnesium sulphate	0.400
Calcium chloride	0.004
Cobalt sulphate	0.001
Cupric sulphate	0.001
Ferrous sulphate	0.001
Zinc sulphate	0.001
Manganese chloride	0.002
Resazurin	0.001
Agar	0.750
Final pH (at 25°C)	7.1±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 40 grams in 1000 ml distilled water containing 3 grams polysorbate 80 and 5 grams glycerol. Heat to boiling to dissolve the medium completely. Dispense as desired and sterilize by autoclaving at 118°C for 15 minutes. Place in cool dark place till use. DO NOT RESTERILIZE the medium

Note: If more than upper one-third of the medium has acquired a pink colour, the medium may be restored once by heating in a water bath or in free flowing steam until the pink colour disappears.

Principle And Interpretation

Clausen Medium was developed by Clausen (1). This medium is also called as HS-T (Dithionite Thioglycollate) Medium and is recommended for sterility testing by the Nordic Pharmacopoeia Board. Random sample selection is recognized by the Board and they refer to the process as microbial-contamination test. The Standard microbial contamination test is developed to establish the number of non-sterile units, if any in batch, is below a specific level. Random sampling in sufficient quantity of the bulk should be examined.

In the microbial contamination test for detecting the non-sterile units, two methods can be used viz. Membrane filter method and Dilution method. The test must be performed with all precautions taken to prevent laboratory contamination.

This medium is very nutritious consisting of casein enzymic hydrolysate, papaic digest of soyabean meal, yeast extract and dextrose. L-cystine and sodium thioglycollate act as reducing agents, and the essential metals help for isolating anaerobic spore-formers. Polysorbate 80 and lecithin are added in this medium to overcome the effects of cationic agents, which can exert

bacteriostatic effect in vitro. This medium is clear in appearance and yellow coloured. Under aerobic conditions it turns pink. Therefore at the time of use the upper one third of the medium should be pink.

The standard microbial contamination test is passed if growth is not observed in any of the tubes. Growth is examined by the appearance of turbidity in fluid or semi fluid media and by the formation of colonies on solid media.

Quality Control

Appearance
Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium
Light straw coloured, clear to slightly opalescent solution with upper 10% or less portion pink on standing.

Reaction
Reaction of 4% w/v aqueous solution containing 0.3% w/v polysorbate 80 and 0.5% w/v glycerol pH : 7.1±0.2

pH
6.90-7.30

Cultural Response
M552: Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours.

Organism	Inoculum (CFU)	Growth
Bacillus subtilis ATCC 6633	50-100	luxuriant
Candida albicans ATCC 10231	50-100	luxuriant
Clostridium sporogenes ATCC 11437	50-100	luxuriant
Pseudomonas aeruginosa ATCC 27853	50-100	luxuriant
Staphylococcus aureus ATCC 25923	50-100	luxuriant
Staphylococcus epidermidis ATCC 12228	50-100	luxuriant
Streptococcus pyogenes ATCC 19615	50-100	luxuriant

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. Clausen O.G., 1973, Pharmaceutica Acta Helvetiae, 48:541.

Revision : 2 / 2015