Gluconate Test Medium

M483

Gluconate Test Medium is used for detecting gluconate-oxidizing microorganisms.

Composition**

Ingredients	Gms / Litre
Casein peptone	1.500
Yeast extract	1.000
Dipotassium hydrogen phosphate	1.000
Potassium gluconate	40.000
Final pH (at 25°C)	7.0±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 43.5 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Dispense 2 ml in screw cap bottles. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Principle And Interpretation

Gluconate Test Medium is used in detection of gluconate oxidizing microorganisms. This medium is used to check the ability of an organism to oxidize gluconates, the sole carbon source, to the reducing compound 2-keto-gluconate which subsequently accumulates in the medium (1).

Casein peptone and yeast extract provides nitrogen and other nutrients necessary to support bacterial growth. Dipotassium hydrogen phosphate buffers the medium. The basis of the test is the change from gluconate, (a non-reducing compound) to 2-keto-gluconate (a reducing compound), which is tested using a suitable reagent (Benedicts reagent). A 4% w/v solution of potassium salt of gluconate is used since at the end of 48 hours of incubation, this amount permits *Pseudomonas aeruginosa* to accumulate at least 50% of potassium 2-ketogluconate (2).

Gluconate Test: Inoculate the medium with the growth from an 18-24 hours pure culture (e.g. Kligler Iron Agar (M078) or Triple Sugar Iron Agar (M021)) and incubate at 37°C for 48 hours. Then add 1 ml of Benedicts reagent for reducing sugars, place the tube in boiling water bath for 10 minutes and observe for the production of a coloured precipitate of cuprous oxide.

Positive: green to orange precipitate

Negative: the blue colour of the reagent is unchanged

Quality Control

Appearance
Off-white to light yellow homogeneous free flowing powder

Colour and Clarity of prepared medium
Light straw coloured, clear solution

Reaction
Reaction of 4.35% w/v aqueous solution at 25°C. pH : 7.0±0.2

pH
6.80-7.20

Cultural Response
M483: Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours.

Organism	Inoculum (CFU)	Growth	Gluconate test
Cultural Response			
Escherichia coli ATCC 25922	50-100	luxuriant	negative, no colour change,

			medium remains blue or bluish green
Pseudomonas aeruginosa ATCC 27853	50-100	luxuriant	positive, yellow to orange red precipitate
Citrobacter freundii ATCC 8090	50-100	luxuriant	negative, no colour change, medium remains blue or bluish green
Klebsiella pneumoniae ATCC 13883	50-100	luxuriant	positive, yellow to orange red precipitate

Storage and Shelf Life

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

Reference

1.Collee J. G., Fraser A. G., Marmion B. P., Simmons A., (Eds.), Mackie and McCartney, Practical Medical Microbiology, 1996, 14th Edition, Churchill Livingstone.
2.Hynes W. C., 1951, J. Gen. Microbiol., 5: 939.

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