

Technical Information

Gluconate Test Medium

Product Code: DM 1483

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

Composition**

Ingredients	Gms / Litre
Peptic digest of animal tissue	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

Principle & Interpretation

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⁾.

Peptic digest of animal tissue 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 to detect the change of gluconate, (a non-reducing compound) to 2-keto-gluconate (a reducing compound), which is tested using a suitable reagent (Benedict's 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 (DM1078) or Triple Sugar Iron Agar (DM1021) and incubate at 37°C for 48 hours. Then add 1 ml of Benedict's 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: the blue colour of reagent changed from blue to green/yellow/orange/precipitates.

Negative: the blue colour of the reagent is unchanged

Methodology

Suspend 43.5 grams of powder media in 1000 ml distilled water. Shake well & heat to dissolve the medium completely. Dispense 2 ml in screw cap bottles. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Quality Control

Physical 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 range 6.80-7.20

Cultural Response/ characteristics

DM 1483: Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours.

Organism	Inoculum (CFU)	Growth	Gluconate test
<i>Escherichia coli</i> ATCC 25922	50-100	luxuriant	negative, no colour change, medium remains blue or bluish green
<i>Pseudomonas aeruginosa</i> ATCC 27853	50-100	luxuriant	positive, yellow to orange red precipitate
<i>Citrobacter freundii</i> ATCC 8090	50-100	luxuriant	negative, no colour change, medium remains blue or bluish green
<i>Klebsiella pneumoniae</i> ATCC 13883	50-100	luxuriant	positive, yellow to orange red precipitate

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° in sealable plastic bags for 2-5 days.

Further Reading

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

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