

OF Glucose Medium (i23891)

A semisolid medium used for the identification of gramnegative bacteria on the basis of their ability to oxidize or ferment glucose Industry: Food

Principles & Uses

This culture medium, used for determining the metabolic processes of carbohydrates by bacteria, differentiates between oxidative and fermentative metabolism. Bacteria metabolize carbohydrates either aerobically (in the presence of atmospheric oxygen) or anaerobically (in the absence of atmospheric oxygen). The medium employs a pH indicator, bromothymol blue, which turns yellow when acid is produced during carbohydrate degradation. The high carbohydrate concentration prevents the utilization of peptones by aerobic microorganisms, avoiding alkaline reactions. Dipotassium phosphate acts as a pH buffer and promotes fermentation, while agar concentration determines motility and aids acid distribution throughout the medium. This medium can be used with various carbohydrates to assess the oxidativefermentative reactions of microorganisms.

Hugh Leifson Medium, designed by Hugh and Leifson, serves to differentiate bacteria based on their fermentation and oxidation of carbohydrates.

Inoculate both aerobic and anaerobic fermentation tubes. In one of the duplicate tubes, create a barrier by adding a 25 mm-thick layer of sterile paraffin oil on the agar surface. Incubate them at 37°C. Oxidative microorganisms generate acid in the unsealed tube with little or no growth, and there is no acid formation in the sealed tube. In contrast, fermentative microorganisms produce acid in both sealed and unsealed tubes. The presence of acid is indicated by a color change throughout the medium, transitioning from greenish blue to yellow. Glucose OF Medium confirms *Enterobacteriaceae* colonies in various samples by testing for glucose fermentation and oxidase presence. This medium is crucial in food and environmental testing and is recommended by ISO 21528 for confirming presumptive *Enterobacteriaceae* colonies through a fermentation test.

Composition (gr/L)

Enzymatic Digest of Casein 2, Glucose 10, Sodium Chloride 5, Dipotassium Hydrogen Phosphate 0.3, Bromothymol Blue 0.08, Agar 3. Final pH at 25° C 6.8 ± 0.2

Preparation from dehydrated Powder

Suspend 20.38 g of the powder in 1 Liter of distilled or deionized water. Mix thoroughly. Dispense and autoclave at 121°C for 15 minutes. Immediately prior to inoculation, heat the tubes, particularly those designated for the fermentative test, in boiling water or a stream of flowing steam for a duration of 15 minutes. This process effectively eliminates any oxygen present. Afterward, swiftly cool the tubes to the required incubation temperature.

Quality Control

Dehydrated Appearance: Beige to orange, freeflowing, homogeneous.

Prepared Appearance: Green, slightly opalescent. Reaction of 2% Solution at 25°C: pH 6.8 ± 0.2

Cultural Response

Inoculate and incubate at $35 \pm 2^{\circ}$ C for 18-24 hours.

Organism (ATCC*)	Recovery	Fermentation Test
Escherichia coli (25922)	Good	+/Yellow
Pseudomonas aeruginosa (27853)	Good	-/Yellow at the top

*ATCC is a registered trade mark of the American Type Culture Collection.





The medium is prepared in tube Number 1, indicated by the green color.

In tubes 2 and 3, *E. coli* demonstrates positive fermentation with a yellow color observed in both tubes, signifying fermentative activity.

Tubes 4 and 5 contain *P. aeruginosa*, which exhibits negative fermentation. In the unsealed tube (4), it appears yellow at the top, while in the sealed tube (5), there is no observable change.

Storage

Keep the container at 15-30 °C. Store prepared medium at 2-8 °C.