OF Basal Medium (Hugh And Leifson)

Cat. 1500

For the identification of non-fermenting bacilli of medical and sanitary importance

Practical information

Aplications	Categories
Differentiation	Non fermentative gram negative bacteria

Industry: Clinical

Principles and uses

OF Basal Medium (Hugh And Leifson) is a semisolid medium, prepared according to Hugh and Leifson's formula, and is used to determine the metabolism (oxidation, fermentation) of Gram-negative bacteria. It is useful for Pseudomonas, Salmonella, Shigella and Alcaligenes.

Casein peptone provides nitrogen, vitamins, minerals and amino acids essential for growth. Bromothymol blue is the pH indicator. Sodium chloride supplies essential electrolytes for transport and osmotic balance. Dipotassium phosphate acts as a buffer system. Bacteriological agar is the solidifying agent.

Formula in g/L

Bromthymol blue	0,03	Bacteriological agar	2,5
Casein peptone	2	Dipotassium phosphate	0,3
Sodium chloride	5		

Preparation

Suspend 9,8 grams of the medium in one litre of distilled water. Heat with frequent agitation until dissolved. Sterilize in autoclave at 121 ºC for 15 minutes. Add 10 ml of 10% glucose (or any suitable sugar) solution, sterilized by filtration, to 100 ml of liquid medium. Mix and aseptically dispense 5 ml per tube. If preferred, add 1,0 grams of carbohydrate directly to 100 ml. of medium and sterilize in the autoclave at 118 ºC for 10 minutes to avoid the degradation of the sugar.

Instructions for use

-Inoculate 2 fresh tubes by stabbing with a fresh culture of the organism in study. If the medium has been prepared and stored, remelt in a water bath to expel the dissolved gases.
-After inoculation, add a layer of 4 to 5 mm of paraffin oil to one of the tubes. It is not recommended to use mineral oil.
-Incubate both tubes at 35°C for 48 hours or more, up to 7 days with the caps loose. -To facilitate the identification of Gram-negative non-fermenting bacilli, also use Indole Nitrate Medium (Cat. 1504).

Results:
1) Fermentation: Yellow color in both tubes with or without gas.
2) Oxidation: Yellow color only in tube without oil.
3) No oxidation/fermentation: No change in the color of the tubes. The carbohydrates have not been fermented or oxidized. Inert microorganisms, e.g. Alcaligenes faecalis.

Quality control

Solubility	Appareance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25ºC)
w/o rests	Fine powder	Beige with a greenish tint	Green-bluish	7,1 ± 0,2

Microbiological test

Incubation conditions: (35±2 ºC / 48-72 h)

K = Alkaline, green (without change)
A = Acid, yellow
G = Gas, sometimes perceptible

Microrganisms	Specification	Characteristic reaction with sacrose	Characteristic reaction w/o sugar	Characteristic reaction with glucose	Characteristic reaction with lactose
Shigella flexneri ATCC 12022	Good growth	Opened: K Closed: K	Abierto: K Cerrado: K	Opened: A Closed: A	Opened: K Closed: K
Salmonella enteritidis ATCC 13076	Good growth	Opened: K Closed: K	Opened: K Closed: K	Opened: AG Closed: AG	Opened: K Closed: K
Escherichia coli ATCC 25922	Good growth	Opened: K Closed: K	Opened: K Closed: K	Opened: AG Closed: AG	Opened: AG Closed: AG
Pseudomonas aeruginosa ATCC 27853	Good growth	Opened: K Closed: K	Opened: K Closed: K	Opened: K Closed: K	Opened: K Closed: K
Alcaligenes faecalis ATCC 8750	Good growth	Opened: K Closed: K	Opened: K Closed: K	Opened: K Closed: K	Opened: K Closed: K

Storage

Temp. Min.:2 ºC
Temp. Max.:25 ºC

Bibliography

Hugh, R. and Leifson, E.J. Bact. 66:24-26. 1953. Lisenko J. Gen. Microbiol., 35:379, 1961. Edwards y Ewing Identification of Enterobacteriaceae. Burguess Publ. Co. Minneapolis, Minn., 1972.