Bushnell Haas Agar

M349

Bushnell Haas Agar is recommended for the microbial examination of fuels and for studying microbial hydrocarbon deterioration.

Composition**

Ingredients	Gms / Litre
Magnesium sulphate	0.200
Calcium chloride	0.020
Monopotassium phosphate	1.000
Dipotassium phosphate	1.000
Ammonium nitrate	1.000
Ferric chloride	0.050
Agar	20.000
Final pH (at 25°C)	7.0±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 23.27 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. A white precipitate prior to sterilization becoming yellow to orange after sterilization is normal.

Principle And Interpretation

Bushnell Haas Agar is prepared as per the formula of Bushnell and Haas (1) and recommended for the microbiological examination of fuels by the SIM Committee on microbiological deteriorations of fuels (2). These media contain all nutrients except carbon source, necessary for the growth of bacteria. Only those bacteria that are able to decompose hydrocarbon will grow in these media. Specific carbon source i.e. hydrocarbon can be added to this medium and their utilization by different microorganisms can be studied.

These bacteria can decompose a variety of hydrocarbons like kerosene, mineral oil, paraffin wax and gasoline. For liquid hydrocarbon the hydrocarbon is layered on the surface of inoculated agar. For testing volatile hydrocarbons such as gasoline the Petri-plates containing the medium are inverted and the hydrocarbon is poured into the lid. Magnesium sulphate, calcium chloride and ferric chloride provide trace elements. Ammonium nitrate is a nitrogen source while monopotassium phosphate and potassium phosphate buffers the medium.

Quality Control

Appearance
White to cream homogeneous free flowing powder

Gelling
Firm, comparable with 2.0% agar gel.

Colour and Clarity of prepared medium
Light amber coloured, clear to slightly opalescent gel forms in Petri plates.

Reaction
Reaction of 2.33% w/v aqueous solution at 25°C. pH : 7.0±0.2

pH
6.80-7.20

Cultural Response
M349: Cultural characteristics observed after an incubation at 25-30°C within 1 week.

Organism	Inoculum (CFU)	Growth (Plain)	Growth w/ minerals
Cultural Response			

Pseudomonas aeruginosa ATCC 27853	50-100	poor	good-luxuriant
Pseudomonas aeruginosa ATCC 9027	50-100	poor	good-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. Bushnell and Haas, 1941, J. Bacteriol., 41:653.
2. Allred, DeGray, Edwards, Hedrick, Klemme, Rogers, Wulf and Hodge, 1963, Proposed Procedures for Microbiological Examination of Fuels, SIM Special Publications, No. 1. Merck, Sharp & Dohme Research Laboratories, Rahway, N.J.

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