



Dehydrated Culture Media
Bases / Media Supplements

Technical Information

H Broth

Product Code: DM 1243

Application: - H Broth is recommended for the preparation of H antigen used in the identification and differentiation of *Salmonella* species.

Composition**

Ingredients	Gms / Litre
Ingredients	5.000
Casein enzymic hydrolysate	5.000
Peptic digest of animal tissue	3.000
Beef extract	1.000
Dextrose	2.500
Dipotassium phosphate	5.000
Sodium chloride	7.2±0.2

Final pH (at 25°C)

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

The genus *Salmonella* is a member of the family *Enterobacteriaceae*. *Salmonella* has three kinds of major antigen namely Somatic (O) or Cell Wall Antigens, Surface (Envelope) Antigens and Flagellar (H) Antigens⁽¹⁾. Identification and differentiation of Salmonellae based on H antigen were found to be less labor-intensive than the standard methods and require no more technical skill. H Broth, used for the preparation of H antigen in the identification and differentiation of Salmonellae, was originally devised by Hajna and Damon⁽²⁾. The medium is also used for differentiating other members of *Enterobacteriaceae* family⁽³⁾. The combination of casein enzymic hydrolysate and peptic digest of animal tissue makes the medium highly nutritive for the growth of gram-negative enteric bacteria⁽⁴⁾.

Peptic digest of animal tissue, casein enzymic hydrolysate and beef extract in the medium provide nitrogen, vitamins and minerals necessary to support bacterial growth. Dextrose is the carbon and energy source. Dipotassium phosphate provides buffering to the medium. Sodium chloride provides essential ions and maintains the osmotic equilibrium.

Presumptive typical colonies should be recovered from SS Agar (DM1108) or Bismuth Sulphite Agar (DM1027) and used to inoculate tubes of TSI Agar (DM1021), Motility Test Medium (DM1260) and H Broth (DM1243). *Salmonella Typhi* or Salmonella-like organisms identified from these tests are subjected to serological testing including indole test.

Methodology

Suspend 21.5 grams of powder media in 1000 ml distilled water. Shake well & heat if necessary to ensure complete solution. Dispense in 4 ml amounts in 13 x 100 mm test tubes and sterilize by autoclaving at 115°C for 15 minutes.



Quality Control

Physical Appearance

Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Light amber coloured, clear solution without any precipitate

Reaction

Reaction of 2.15% w/v aqueous solution at 25°C. pH : 7.2±0.2

pH Range 7.00-7.40

Cultural Response/Characteristics

DM 1243: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth
<i>Salmonella Typhi</i> ATCC 6539	50-100	luxuriant
<i>Salmonella Typhimurium</i> ATCC 14028	50-100	luxuriant
<i>Salmonella Enteritidis</i> ATCC 13076	50-100	luxuriant
<i>Salmonella Paratyphi A</i> ATCC 9150	50-100	
<i>Salmonella Paratyphi B</i> ATCC 8759	50-100	luxuriant
<i>Salmonella Arizonae</i>	50-100	luxuriant

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: 2-8⁰ in sealable plastic bags for 2-5 days.

Further Reading

1. Gruenewald R., Dixon D. P., Brun M., Yappow S., Henderson R., Douglas J. E., and Backer M. H., Appl. Environ. Microbiol., 1990, 56 (1),24-30
2. Hajna A. A. and Damon S. R., 1950, Pub. Health Rep., 65:116,
3. Hajna A. A., 1951, Pub. Health Lab., 9:23,
4. Hajna A. A., 1951, Personal communication.

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