

Hektoen Enteric Agar ISO

Cat. 1030

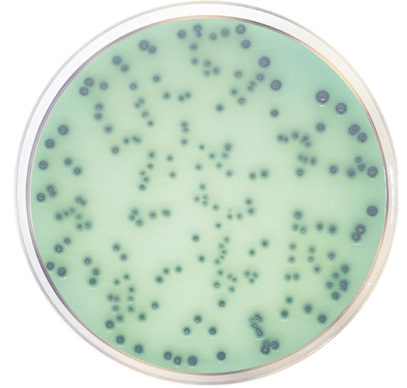
For the isolation and differentiation of Gram negative enteric bacteria

Practical information

Applications	Categories
Selective isolation	Salmonella
Selective isolation	Shigella
Differentiation	Enterobacteria

Industry: Clinical / Food

Regulations: ISO 21567



Principles and uses

Hektoen Enteric Agar is a differential and selective medium used for isolating and differentiating enteric pathogens such as Salmonella and Shigella, both of which cause a variety of serious human gastrointestinal diseases; and other Gram-negative Enterobacteriaceae.

It is used particularly in foods where multi-steps are followed to isolate the pathogens of gastroenteritis. The nutrients for growth are provided by the Meat Peptone and Yeast extract. The increased content of the Peptone and the three fermentable carbohydrates (Lactose, Sucrose, Salicin) as sources of carbon and energy reduce the inhibitory action of the Bile salts on Salmonella and Shigella spp. The lactose concentration in this medium is higher than in many other media used for enterics since this helps the visualization of enteric pathogens and minimizes the problem of delayed lactose fermentation. Bromothymol blue and Acid fuchsin are pH indicators. Sodium thiosulfate provides Sulphur, and Ferric ammonium citrate is the indicator for H₂S production. H₂S positive colonies are black-centered. Sodium chloride maintains the osmotic balance.

The norma ISO 21567 recommends the Hektoen Agar as a selective solid media for the enumeration of Shigella spp.

Although suppressed, partially inhibited E. coli and other organisms which use lactose, sucrose, and/or salicin with the production of acid, give colonies whose tones vary from yellow to orange to salmon. The Salmonella and Shigella are green or green-blue. Proteus is not inhibited but produces a green-yellow colony when it grows. The colonies of Proteus and Salmonella may present a black center and clear edges if they form iron sulfide as a result of H₂S production.

Formula in g/L

Bromthymol blue	0,065	Acid fuchsin	0,1
Bacteriological agar	14	Bile salts N° 3	9
Ferric ammonium citrate	1,5	Lactose	12
Salicin	2	Sodium chloride	5
Sodium thiosulfate	5	Sucrose	12
Yeast extract	3	Enzymatic digest of meat	12

Typical formula g/L * Adjusted and/or supplemented as required to meet performance criteria.

Preparation

Suspend 75,6 grams of the medium in one liter of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. AVOID OVERHEATING. DO NOT AUTOCLAVE. Cool to 47 °C and pour into Petri dishes.

Instructions for use

» For clinical diagnosis, the type of sample is feces.

- Inoculate on the surface. Parallel striae with the handle or swab. The inoculation can also be done from a pre-enrichment culture.
- Incubate at 35±2 °C for 18-24 hours.
- Reading and interpretation of the results.

» For other uses not covered by the CE marking:

Enumeration of *Shigella* spp. according to ISO 21567:

- Seed the specimen by streaking directly on the surface of the medium, or previously enrich in Tetrathionate Broth (Cat. 1114), Selenite Cystine Broth (Cat. 1220), GN Broth (Cat. 1248) or Shigella Broth (Cat. 2078) for the detection of *Shigella*.
- Incubate at 37±1 °C for 20-24 hours.
- It is recommended to seed the sample on other selective media at the same time for Enterobacteriaceae because a larger number of positive cultures will be obtained. These media can be, for example, Eosin Methylene Blue Agar (Cat. 1039), MacConkey Agar (Cat. 1052), SS Agar (Cat. 1064), Brilliant Green Agar (Cat. 1010), Desoxycholate Lactose Agar (Cat. 1025), XLD Agar (Cat. 1080),.

Quality control

Solubility	Appearance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25°C)
w/o rests	Fine powder	Beige	Dusky green	7,5±0,2

Microbiological test

Incubation conditions: (37±1 °C / 20-24 h)

Inoculation conditions: Productivity (10³-10⁴ CFU) / Selectivity (10⁴-10⁶ CFU)

Microorganisms	Specification	Characteristic reaction
<i>Enterococcus faecalis</i> ATCC 11700	Total inhibition	
<i>Shigella flexneri</i> ATCC 12022	Good growth	Blue-green colonies
<i>Klebsiella aerogenes</i> ATCC 13048	Moderate growth	Red-salmon colonies
<i>Salmonella enteritidis</i> ATCC 13076	Good growth	Blue-green colonies
<i>Salmonella typhimurium</i> ATCC 14028	Good growth	Blue-green with black center colonies
<i>Escherichia coli</i> ATCC 25922	Moderate growth	Red-salmon colonies

Storage

Temp. Min.:2 °C
Temp. Max.:25 °C

Bibliography

ISO 21567:2004 Microbiology of food and animal feeding stuffs -- Horizontal method for the detection of *Shigella* spp.
King, S. & Metzger Appl. Microbiol. 16:577. 1968. King, S. & Metzger Appl. Microbiol. 16:579, 1968.
Isenberg, Kominos & Siegel. Appl. Microbiol. 18:656. 1969. Hoben, Aston & Peterson Appl. Microbiol. 26:126. 1973.
Polloch & Dalhgren. Appl. Microbiol. 27:197. 1974. Peloxv, Laviotte & Pons Microbia, Tomo I No. 1. 1975.
Goo et al Appl. Microbiol. 26:288, 1973.