Hektoen Enteric (HE) Agar CE (NCM2021)

Intended Use
Hektoen Enteric Agar is used for the isolation and differentiation of enteric pathogens.

Description
Hektoen Enteric Agar was developed in 1967 by King and Metzger. Compared to other enteric differentiating media commonly used, Hektoen Enteric Agar increased the isolation rate of *Salmonella* spp. and *Shigella* spp. This was accomplished by increasing the carbohydrate and peptone content of the medium in order to counteract the inhibitory effects of bile salts and indicators. King and Metzger formulated a medium that slightly inhibited growth of *Salmonella* and *Shigella*, while inhibiting Gram-positive microorganisms.

Principles of the Procedure
Enzymatic Digest of Animal Tissue provides nitrogen, carbon, and amino acids required for organism growth. Yeast Extract is a vitamin source. Bile Salts Mixture and Acid Fuchsin inhibit Gram-positive organisms. Lactose, Sucrose, and Salicin are fermentable carbohydrates. Sodium Chloride maintains the osmotic balance of the medium. Ferric Ammonium Citrate, a source of iron, allows the detection of hydrogen sulfide (H_2S) produced from Sodium Thiosulfate. H_2S-positive colonies have black centers. Bromothymol Blue is added as the pH indicator. Agar is the solidifying agent.

Typical Formulation

Meat Peptone	12.0 g/L
Yeast Extract	3.0 g/L
Lactose	12.0 g/L
Sucrose	12.0 g/L
Salicin	2.0 g/L
Bile Salts No. 3	7.0 g/L
Sodium Deoxycholate	2.4 g/L
Sodium Chloride	5.0 g/L
Sodium Thiosulfate	5.0 g/L
Ferric Ammonium Citrate	1.5 g/L
Acid Fuchsin	0.1 g/L
Bromothymol Blue	0.065 g/L
Agar No. 1	14.0 g/L

pH: 7.5 ± 0.2 at 25°C
Formula may be adjusted and/or supplemented as required to meet performance specifications.

Precaution
Refer to SDS

Preparation
1. Disperse 76g in one liter of deionized water.
2. Soak for 10 minutes, swirl to mix and sterilize by bringing to the boil.
3. DO NOT AUTOCLAVE OR OVERHEAT THIS MEDIUM.
4. Cool to 45-50°C and mix before pouring into Petri dishes and dry the agar surface.

Test Procedure
For isolation and identification of pathogenic *Enterobacteriaceae* refer to appropriate references.

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Quality Control Specifications

Dehydrated Appearance: Powder is homogeneous, free flowing, and light greenish beige.

Prepared Appearance: Prepared medium is trace to slightly hazy and light to dark green.

Expected Cultural Response: Cultural response on Hektoen Enteric Agar at 37°C ± 1°C after 24 ± 3 hours incubation.

Microorganism	Growth	Reaction
Escherichia coli ATCC® 25922	Suppressed growth to complete inhibition	yellow to salmon-orange colonies
Enterococcus faecalis ATCC® 29212	Complete inhibition	---
Salmonella enteritidis ATCC® 13076	≥70%	Green w/ black center
Salmonella typhimurium ATCC® 14028	≥70%	Green w/ black center
Shigella sonnei NCTC® 8574	≥50%	Green colonies

Results

Refer to appropriate references and procedures for results.

Expiration

Refer to expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitations of the Procedure

1. Do not autoclave medium because excessive heat may alter ingredients.
2. *Proteus* spp. may resemble salmonellae or shigellae. Further testing should be conducted to confirm the presumptive identification or organisms isolated on this medium.
3. Due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.

Storage

Store dehydrated culture media at 2–30°C away from direct sunlight. Once opened and recapped, place the container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping container tightly closed.

References

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