## **Hektoen Enteric Agar**

Selective and differential medium for detection of pathogenic intestinal bacteria from food and clinical specimens, according to ISO 21567.

### DESCRIPTION

Hektoen Enteric Agar is a moderately selective medium used for the isolation and cultivation of Gram-negative enteric microorganisms, especially *Shigella* spp, from faeces, foodstuffs and other materials of sanitary importance.

This medium meets the requirements of the APHA and ISO 21567 for the isolation and differentiation of *Salmonella* and *Shigella* spp.

TYPICAL FORMULA	(g/ <b>l</b> )
Enzymatic Digest of Meat	12.0
Yeast Extract	3.0
Lactose	12.0
Saccharose	12.0
Salicin	2.0
Bile Salts No. 3	9.0
Sodium Chloride	5.0
Sodium Thiosulfate	5.0
Ammonium Ferric Citrate	1.5
Acid Fuchsin	0.1
Bromothymol Blue	0.065
Agar	15.0
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Final pH  $7.5 \pm 0.2$  at 25°C

## **METHOD PRINCIPLE**

Enzymatic digest of meat provides amino acids, nitrogen, carbon, vitamins and minerals for organisms growth. Yeast extract is a source of vitamins, particularly of B-group. Lactose, saccharose and salicin are fermentable carbohydrates. Bile salts and acid fuchsin inhibit Gram-positive organisms. Sodium chloride maintains the osmotic balance of the medium. Ammonium ferric citrate and sodium thiosulfate enable the detection of hydrogen sulfide production. Bromothymol blue together with acid fuchsin act are the pH indicator system. Agar is the solidifying agent.

#### **PREPARATION**

Dehydrated medium	Suspend 76 g of th	e powder in	1 liter of distilled or deionized water. Mix well. Heat to boil

shaking frequently until completely dissolved. DO NOT AUTOCLAVE.

# Medium in bottles Melt the content of the bottle in a water bath at 100°C (loosing the cap partially removed) until

completely dissolved. Then screw the cap and check the homogeneity of the dissolved medium, if it is the case turning the bottle upside down. Cool at 45-50°C, mix well avoiding foam formation and aseptically distribute into Petri dishes.

## **TEST PROCEDURE**

Inoculate the plates by directly streaking the specimen on the agar surface or spread the sample from an enrichment culture. Incubate aerobically at  $35 \pm 2$ °C for 18-24 h

### **INTERPRETING RESULTS**

Shigella and Providencia spp, form green, moist colonies.

Salmonella and Proteus spp, grow as blue-green colonies, with or without black center due to H<sub>2</sub>S production.

Coliforms, which are mostly rapid lactose-saccharose-salicin fermenters, develop red-salmon colonies surrounded by a zone of bile precipitate.

Enterococci, Staphylococci and other Gram-positive bacteria are partially or completely inhibited.

Notice that further testing should be conducted to confirm the presumptive identification of organisms isolated on this medium.

### APPEARANCE OF THE MEDIUM

Dehydrated medium: free-flowing, homogeneous, light beige. Prepared medium: slightly opalescent, green.

## **STORAGE**

The powder is very hygroscopic, store the powder at 10-30°C, in a dry environment, in its original container tightly closed. Store bottles and prepared plates at 10-25°C away from light. Do not use the product beyond its expiry date on the label or if product shows any evidence of contamination or any sign of deterioration.

## SHELF LIFE

Dehydrated medium: 4 years. Medium in bottles: 2 years. Ready-to-use plates: 6 months.

Distribué par :

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## **QUALITY CONTROL**

Plates are inoculated with the microbial strains indicated in the QC table.

Inoculum for productivity: 50-100 CFU Inoculum for selectivity: 10<sup>4</sup>-10<sup>6</sup> CFU

Incubation conditions: aerobically at  $35 \pm 2^{\circ}$ C for 18-24 hours.

#### QC Table.

Microorganism		Growth	Specification
Salmonella Typhimurium	ATCC® 14028	Good	Blue-green colonies with black centre
Shigella flexneri	ATCC® 12022	Good	Green colonies
Proteus mirabilis	ATCC® 12453	Good	Blue-green colonies with black centre
Klebsiella pneumoniae	ATCC® 13883	Good	Red-salmon colonies with zone of bile precipitate
Escherichia coli	ATCC® 8739	Partially to completely inhibited	Red-salmon colonies with or without zone of bile precipitate
Enterococcus faecalis	ATCC® 29212	Inhibited	

#### WARNING AND PRECAUTIONS

The product does not contain hazardous substances in concentrations exceeding the limits set by current legislation and therefore is not classified as dangerous. It is nevertheless recommended to consult the safety data sheet for its correct use. The product is intended for *in vitro* diagnostic use only and must be used by properly trained operators.

#### DISPOSAL OF WASTE

Disposal of waste must be carried out according to national and local regulations in force.

## **BIBLIOGRAPHY**

- ISO 21567:2004. Microbiology of food and animal feeding stuffs -- Horizontal method for the detection of Shigella spp.
- Perez J.M., P. Cavalli, C. Roure, R. Renac, Y. Gille, and A. M. Freydiere (2003) Comparison of Four Chromogenic Media and Hektoen Agar for Detection and Presumptive Identification of Salmonella Strains in Human Stools. J Clin Microbiol; 41(3):1130–1134.
- American Public Health Association (1992) Compendium of Methods for the Microbiological Examination of Foods 3rd Edition. APHA Inc. Washington DC.
- Bisciello N.B. jr. and Schrade J.(1974) Evaluation of Hektoen Enteric Agar for the detection of Salmonella in foods and feeds. Journ of AOAC; 57: 992-996.

PRESENTATION		Contents	Ref.
Hektoen Enteric Agar	90 mm ready-to-use plates	20 plates	10043
Hektoen Enteric Agar	90 mm ready-to-use plates	100 plates	10043*
Hektoen Enteric Agar	Bottles	6 x 100 ml bottles	402230
Hektoen Enteric Agar	Bottles	6 x 200 ml bottles	412230
Hektoen Enteric Agar	Dehydrated medium	500 g of powder	610021
Hektoen Enteric Agar	Dehydrated medium	100 g of powder	620021
Hektoen Enteric Agar	Dehydrated medium	5 kg of powder	6100215

TABLE OF SYMBOLS			
LOT Batch code	IVD In vitro Medical Diagnostic Device	Manufacturer Use by	Fragile, handle with care
REF Catalogue number	Temperature limitation	Contains sufficient for <a href="#">Contains sufficient for <a href="#">Caution, consult Instruction For Use</a></a>	Do not reuse

