

CM 20799 - HEKTOEN ENTERIC AGAR MEDIUM (as per USP)

INTENDED USE

For differential and selective isolation of Salmonella and Shigella species from enteric pathological specimens.

PRODUCT SUMMARY AND EXPLANATION

Hektoen Enteric Agar, a selective and differential medium designed to isolate and differentiate members of the species Salmonella and Shigella from other Enterobacteriaceae and was developed by King and Metzger. When compared with other selective medium, this medium inhibits the growth of Salmonella and Shigella very slightly; thus giving high yields of these microorganisms, but at the same time inhibits accompanying gram positive and other microorganisms. This medium is recommended by United States Pharmacopoeia, 2009 for testing the presence of Salmonella in dietary supplements. This medium is recommended in testing of Salmonella in food sample by various standards. Compared to other differentiating media commonly used in clinical laboratories, Hektoen Enteric Agar is efficient in increasing the isolation rate of Salmonella sp.

Enterobacters that are capable of fermenting one or more of the carbohydrates produces yellow or salmon-orange coloured colonies like Klebsiella pneumonia. that ferments lactose. Non-fermenters will produce blue-green colonies. Organisms that reduce sulfur to hydrogen sulfide will produce black colonies or blue-green colonies with a black center. Salmonella reduce sulfur to hydrogen sulfide, producing a black precipitate. Micrococcus luteus does not grow.

COMPOSITION

Ingredients	Gms / Ltr
Protease peptone	12.000
Yeast extract	3.000
Lactose	12.000
Sucrose	2.000
Salicin	9.000
Bile Salts mixture (Equivalent to Bile Salt No. 3)	9.000
Sodium chloride	5.000
Sodium thiosulfate	5.000
Ferric ammonium citrate	1.500
Acid fuchsin	0.100
Bromothymol blue	0.065
Agar	14.000

PRINCIPLE

Bile salts, bromthymol blue and acid fuchsin inhibit the growth of most Gram positive organisms. Lactose, salicin and sucrose, serves as fermentable source of carbohydrates to encourage the growth and differentiation of enteric bacteria. In this medium by increasing the carbohydrate and peptone content of the medium the inhibitory effect of bile salts and indicators are countered. Protease peptone provides nitrogen, carbon, and amino acids required for organism growth. Yeast Extract is a vitamin source. Sodium chloride maintains the osmotic balance of the medium. Sodium thiosulfate provides a source of sulfur. Hektoen Enteric Agar can also detect the production of hydrogen sulfide gas, which turns parts of the medium black. Ferric ammonium citrate serves as iron source, which cause production of hydrogen sulfide from sodium thiosulphate and also aids in the visualization of hydrogen sulfide production by reacting with hydrogen sulfide gas to form a black precipitate.



INSTRUCTION FOR USE

Dissolve 72.66 grams in 1000 ml purified/ distilled water.
Heat to boiling to dissolve the medium completely, do not autoclave.
Cool to 45-50°C. Mix well and pour into sterile Petri plates.

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder : Cream to yellow with tancast homogeneous free flowing powder.
Appearance of prepared medium : Green coloured clear to slightly opalescent gel forms in Petri plates.
pH (at 25°C) : 7.5±0.2

INTERPRETATION

GrowthPromotionwas observed in accordance with USP, after an incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Colour of Colony	Incubation Temperature	Incubation Period
Salmonella Typhimurium	14028	50-100	Luxuriant	≥70 %	Blue-green with or without black centers	30-35°C	24-48 Hours
Salmonella Abony	6017	50-100	Luxuriant	≥70 %	Blue-green with or without black centers	30-35°C	24-48 Hours
Salmonella Enteritidis	13076	50-100	Luxuriant	≥70 %	Blue-green with or without black centers	30-35°C	24-48 Hours
Salmonella Typhi	6539	50-100	Fair-good	20-40 %	Blue-green with or without black centers	30-35°C	24-48 Hours
Escherichia coli	25922	50-100	None-poor	0-10 %	Orange (may have bile precipitate)	30-35°C	24-48 Hours
Escherichia coli	8739	50-100	None-poor	0-10 %	Orange (may have bile precipitate)	30-35°C	24-48 Hours
Shigella flexneri	12022	50-100	Luxuriant	≥70 %	Greenish blue	30-35°C	24-48 Hours
Enterococcus faecalis	29212	≥10 ³	Inhibited	0%	-	30-35°C	24-48 Hours



Staphylococcus aureus	6538	$\geq 10^3$	Inhibited	0%	-	30-35°C	24-48 Hours
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PACKAGING:

In pack size of 100 gm and 500 gm bottles.

STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

REFERENCES

1. King, S., and W. I. Metzger. 1968. Appl. Microbiol. 16:577
2. King, S., and W. I. Metzger. 1968. Appl. Microbiol. 16:579
3. United States Pharmacopoeia 2009, US Pharmacopoeial Convention, Inc., Rockville, MD
4. Marshall, R. T. (ed.). 1993. Standard methods for the microbiological examination of dairy products, 16th ed. American Public Health Association, Washington, D.C.
5. Downes F P and Ito K (Eds.), 2001, Compendium of Methods for The Microbiological Examination of Foods, 4th ed., APHA, Washington, D.C
6. AOAC, 2005, Bacteriological Analytical Manual, 18th ed., AOAC, Washington, DC

 Good Manufacturing Practices Certified	 For In Vitro Diagnostic Use	 Quantity	 Lot / Batch Number	 Catalogue Number	 Manufacturer
 Temperature Unit	 Authorized Representative MedNet GmbH Buckstrasse 10, 49163 Mander, Germany	 European Conformity	 QR Code	 Consults Instructions for Use	 Best Before

NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.

*For Lab Use Only

