

TM 2108 - HEKTOEN ENTERIC AGAR, W/ 1.4% AGAR

INTENDED USE

For the differential isolation of *Shigella* and *Salmonella* from food specimens in accordance with FDA BAM, 1998.

PRODUCT SUMMARY AND EXPLANATION

Hektoen Enteric Agar w/ 1.4% agar is used for the differential isolation of *Salmonella* and *Shigella* from food specimens in accordance with FDA BAM, 1998. Foods containing poultry, eggs or dairy products are the most frequent vehicles for foodborne Salmonellosis, and a variety of procedures have been developed using Hektoen Enteric Agar as part of the multi-step procedure to isolate *Salmonella*.

Grow suspected sample in Tetrathionate broth overnight. Mix (vortex, if tube) and streak 3 mm loopful (10 µl) incubated TT broth on Hektoen Enteric Agar w/ 1.4% agar. Appearance of blue-green to blue colonies with or without black centers indicates the presence of *Salmonella*. Many cultures of *Salmonella* may produce colonies with large, glossy black centers or may appear as almost completely black colonies.

COMPOSITION

Ingredients	Gms / Ltr
Peptone	12.000
Yeast extract	3.000
Sodium Chloride	5.000
Bile salt mixture	9.000
Lactose	12.000
Sucrose	12.000
Salicin	2.000
Sodium thiosulphate	5.000
Ferric ammonium citrate	1.500
Bromothymol blue	0.065
Acid fuchsin	0.100
Agar	14.000

PRINCIPLE

The increased concentration of carbohydrates and peptic digest of animal tissue helps to reduce the inhibitory effect of bile salts and indicators thus allowing the good growth of *Salmonella* and *Shigella* species while inhibiting the normal intestinal flora. The medium contains three carbohydrates i.e. lactose, sucrose and salicin for differentiation of enteric pathogens. The higher lactose concentration aids in the visualization of enteric pathogens and minimizes the problem of delayed lactose fermentation. Salicin is fermented by many coliforms including those that do not ferment lactose and sucrose. Combination of ferric ammonium citrate and sodium thiosulphate in the medium enables the detection of hydrogen sulfide production indicated by the black coloured colonies. The indicator system, consisting of acid fuchsin and bromothymol blue, has lower toxicity as compared to other enteric media, resulting in improved recovery of enteric pathogens. Low concentration of bile salts allows the growth of *Shigella* and *Salmonellae*. Higher concentration of peptone minimizes the inhibitory effects of the bile salts.

INSTRUCTION FOR USE

- Dissolve 75.67 grams in 1000 ml distilled water.

- Heat to boiling to dissolve the medium completely, do not autoclave or overheat.
- 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

Cultural characteristics observed after an incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Colour of colony	Incubation Temperature	Incubation Period
<i>Escherichia coli</i>	25922	50-100	Fair	20-30%	Orange (may have bile precipitate)	35°C	22-26 Hours
<i>Enterobacter aerogenes</i>	13048	50-100	Fair-good	20-40%	Salmon-orange	35°C	22-26 Hours
<i>Enterococcus faecalis</i>	29212	≥10 ³	Inhibited	0%	-	35°C	22-26 Hours
<i>Salmonella Enteritidis</i>	13076	50-100	Luxuriant	≥70%	Greenish blue may have black centres(H ₂ S production)	35°C	22-26 Hours
<i>Salmonella Typhi</i>	6539	50-100	Luxuriant	≥70%	Greenish blue may have black centres(H ₂ S production)	35°C	22-26 Hours
<i>Salmonella Typhimurium</i>	14028	50-100	Luxuriant	≥70%	Greenish blue may have black centres(H ₂ S production)	35°C	22-26 Hours
<i>Shigella flexneri</i>	12022	50-100	Luxuriant	≥70%	Greenish blue	35°C	22-26 Hours
<i>Escherichia coli</i>	8739	50-100	Fair	20-30%	Orange (may have bile precipitate)	35°C	22-26 Hours

PACKAGING:

In pack size of 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 Metzger W. I., 1967, Abstr. M99, p. 77. Bacteriol. Proc. Am. Soc. Microbiol. 2. King S. and Metzger W. I., 1968, Appl. Microbiol., 16:577.
2. King S. and Metzger W. I., 1968, Appl. Microbiol., 16:579.
3. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. I. Williams & Wilkins, Baltimore, Md.
4. Murray P. R., Baron E. J., Jorgensen J. H., Tenover F. C., Tenover F. C., (Eds.), 8th Ed., 2003, Manual of Clinical Microbiology, ASM, Washington, D.C.
5. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.
6. FDA Bacteriological Analytical Manual, 2005, 18th Ed., AOAC, Washington, DC.
7. Williams, (Ed.), 2005, Official Methods of Analysis of the Association of Official Analytical Chemists, 19th Ed. AOAC, Washington, D.C.
8. Downes F. P. and Ito K., (Ed.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., American Public Health Association, Washington, D.C.
9. Giannella R. A., 1996, Salmonella. In: Barons Medical Microbiology (Baron S et al, eds.), 4th Ed., Univ. of Texas Medical Branch, Hoboken D.A., Ashton D.H.A. and Peterson A.C., 1973, Appl. Microbiol., 21:126.
10. Taylor W.I. and Schelhaut, 1971, Appl. Microbiol., 21:32.

 Good Manufacturing Practices Certified	 Best Before	 Quantity	 Catalogue Number	 Manufacturer
 Temperature Unit	 Lot / Batch Number	 Consults Instructions for Use	 QR Code	

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

***For Lab Use Only**
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