TMP 037 – HEKTOEN ENTERIC AGAR PLATE

INTENDED USE

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

PRODUCT SUMMARY AND EXPLANATION

Hektoen Enteric Agar Plate was developed in 1967 by King and Metzger of the Hektoen Institute in order to increase the frequencies of isolation of Shigella and Salmonella organisms as compared from other media that were frequently utilized in clinical laboratories at that time. In this medium, sodium deoxycholate has been replaced by bile salts in reduced concentration. This allows growth of Shigella as well as the Salmonellae. The peptone concentrations have been increased in order to offset the inhibitory effects of the bile salts. Hektoen Enteric Agar is currently recommended as one of several plating media for the culture of Enterobacteriaceae from stool specimens. 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*. This medium is recommended by United States Pharmacopoeia, 2009 for testing the presence of Salmonella in dietary supplements. This medium is also recommended in testing of *Shigella* in food sample by various standards like ISO 21567.

COMPOSITION

Ingredients	Gms / Ltr
Protease peptone	12.000
Yeast Extract	3.000
Lactose	12.000
Sucrose	12.000
Salicin	9.000
Bile Salt Mixture	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

The increased concentration of carbohydrate and proteose peptone helps to reduce the inhibitory effect of bile salts and indicators and allows 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, thereby aiding in the differentiation process due to the formation of black centered 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.

INSTRUCTION FOR USE

Either streak, inoculate or surface spread the test inoculum aseptically on the plate.

QUALITY CONTROL SPECIFICATIONS

Appearance	:	Green colour, clear to slightly opalescent gel
Quantity of Medium	:	25ml of medium in 90mm plates.
pH (at 25°C)	:	7.5 ± 0.2
Sterility Check	:	Passes release criteria

INTERPRETATION

Cultural response was observed after incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Color of the colony	Incubation Temperature	Incubation Period
Salmonella Typhimurium	14028	50-100	luxuriant	>=70%	Blue green with or without black center	35-37°C	18-24 Hours
Salmonella Enteritidis	13076	50-100	luxuriant	>=70%	Blue green with or without black center	35-37°C	18-24 Hours
Salmonella Typhi	6539	50-100	luxuriant	>=70%	Blue green with or without black center	35-37°C	18-24 Hours
Escherichia coli	25922	50-100	Fair	20-30 %	Orange (May have bile precipitate)	35-37°C	18-24 Hours
Shigella flexneri	12022	50-100	luxuriant	>=70%	Greenish Blue	35-37° C	18-24 Hours
Enterococcus faecalis	29212	$>10^3$	Inhibited	0%	-	35-37°C	18-24 Hours

PACKAGING:

Doubled layered packing containing 5 No. of plates with one silica gel desiccant bag packed inside it.

STORAGE

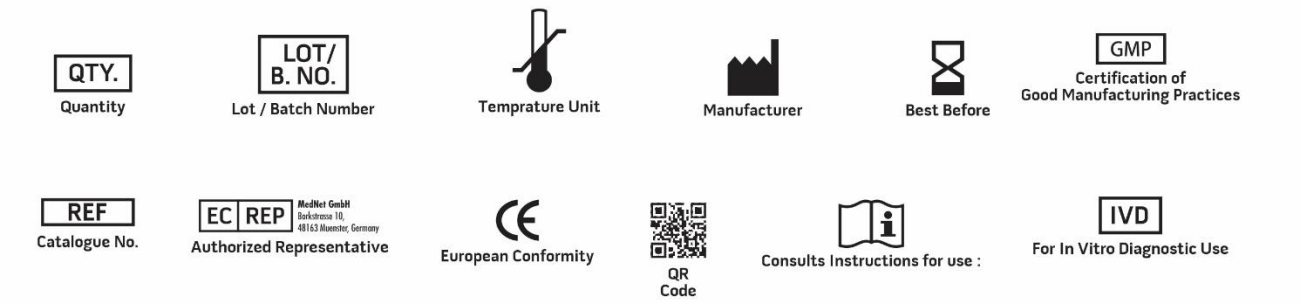
On receipt, store the plates at 15–30 °C. Avoid freezing and overheating. Do not open until ready to use. Prepared plates stored in their original sleeve wrapping until just prior to use may be inoculated up to the expiration date and incubated for recommended incubation times. Allow the medium to warm to room temperature before inoculation.
Product Deterioration: Do not use plates if they show evidence of microbial contamination, discoloration, drying, cracking or 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



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

***For Lab Use Only**

Revision: 22nd March,. 2022