

TMT 017- TRYPTOPHAN MEDIUM

INTENDED USE

For detection of indole production

PRODUCT SUMMARY AND EXPLANATION

Enterohemorrhagic Escherichia coli (EHEC) is a defined subset of Shiga-like (vero) toxin-producing *E. coli*. EHEC infections are waterborne or food borne. EHEC is ingested most commonly with undercooked ground beef. There are more than 50 serotypes of EHEC. However, *E. coli* O157:H7 is the prototype EHEC. *E. coli* O157:H7 can cause an asymptomatic infection, mild diarrhea, or a diarrheal illness that is characterized by non-bloody (progressing to bloody) diarrhea and abdominal cramps (together known as hemorrhagic colitis), few leukocytes in stools and lack of significant fever. Tryptophan Medium is prepared as per the formula approved by ISO Committee, that is a modification of original formula of APHA where the medium is devoid of tryptophan.

This medium is useful for the detection of indole production by *Escherichia coli* O157: H7, which is a key feature in differentiation of coliforms. Certain microorganism breakdown tryptophan with the help of the enzyme tryptophanase that mediate the production of indole by hydrolytic activity. The indole produced can be detected by Kovacs or Ehrlichs reagent. Indole combines with the aldehyde present in the above reagent to give red colour in the alcohol layer. The alcohol layer extracts and concentrates the red colour complex. The test sample is enriched in Modified Soyabean Bile Broth Base by incubating at 42°C for 18-24 hours. *E. coli* O157:H7 is then isolated on MacConkey Sorbitol Agar Base. Pale coloured colonies obtained on incubation at 35-37°C for 18-24 hours are reported as presumptive *E. coli* O157:H7. Presumptive colonies are subjected to indole test that makes the use of Tryptophan Medium.

COMPOSITION

Ingredients	Gms / Ltr
Casein enzymic hydrolysate	10.000
Sodium chloride	5.000
DL-Tryptophan	1.000

PRINCIPLE

Casein enzymic hydrolysate provides carbonaceous and nitrogenous sources required for the growth of microorganisms. Tryptophan is an amino acid, which serves as a substrate to study indole reaction.

INSTRUCTION FOR USE

Inoculate the sample and Incubate at specified temperature and time.

QUALITY CONTROL SPECIFICATION

Appearance of prepared medium	:	Yellow coloured clear solution without any precipitate.
Quantity of Medium	:	10 ml of medium in tubes.
pH (at 25°C)	:	7.5 ± 0.2
Sterility Check	:	Passes release criteria

INTERPRETATION

Cultural characteristics observed after incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Indole production	Incubation Temperature	Incubation Period



<i>Enterobacter aerogenes</i>	13048	50-100	Luxuriant	Negative reaction, no colour development / cloudy ring	35-37°C	18-24 Hours
<i>Escherichia coli</i>	25922	50-100	Luxuriant	Positive reaction, red ring at the interface of the medium	35-37°C	18-24 Hours

PACKAGING:

Pack of 25 Ready-To-Use Liquid Medium tubes containing 10 ml in each tube.

Pack of 50 Ready-To-Use Liquid Medium tubes containing 10 ml in each tube.

STORAGE

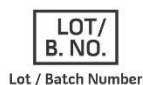
On receipt, store tubes in the dark at 10-25°C. Avoid freezing and overheating. Do not open until ready to use. Minimize exposure to light. Tubed media stored as labeled until just prior to use may be inoculated up to the expiration date and incubated for the recommended incubation times. Allow the medium to warm to room temperature before inoculation.

DISPOSAL

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques.

REFERENCES

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- Murray P. R., Baron J. H., Tenover F. C. and Tenover F. C., (Eds.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.
- International Organization for Standardisation (ISO) Draft: ISO/DIS 16654:1999.
- Eaton A. D., Clesceri L. S., Rice E. W. and Greenberg A. W., (Eds.), 2005, Standard Methods for the Examination of Water and Wastewater, 21st Ed., APHA, Washington, D.C.
- MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore.
- Finegold S. M. and Baron E. J., 1986, Bailey and Scotts Diagnostic Microbiology, 7th Ed., The C.V. Mosby Co., St. Louis.



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

***For Lab Use Only**
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