TMV 468 – TRYPTONE NITRATE MEDIUM (INDOLE NITRATE MEDIUM) (VEG.)

INTENDED USE

For identification of microorganisms by Indole production and Nitrate reduction.

PRODUCT SUMMARY AND EXPLANATION

This medium is prepared by replacing Casein enzymic hydrolysate with Veg hydrolysate which is free of BSE/ TSE risks. This medium can be used against animal based Indole Nitrate medium for identification of microorganisms based on nitrate reduction and indole tests.

Indole production can be detected by the addition of Kovac's Indole reagent or Ehrlich's aldehyde reagent. The formation of a pinkish red colour within 10 seconds in the reagent layer after gentle agitation indicates positive indole test. Potassium nitrate in the medium acts as the substrate for nitrate reduction by microorganisms. Certain bacteria convert nitrate to nitrite, ammonia or nitrogen gas. The presence of nitrite is determined by addition of 0.5 ml of each of Sulphanilic Acid and alpha - Naphthylamine solution. The development of red violet colour indicates nitrate reduction to nitrite. If no colour develops, it means that either nitrate is not reduced or further reduction to ammonia or nitrogen gas has taken place. This can be verified by adding a pinch of zinc dust to the tube. Zinc reduces nitrate to nitrite resulting in a red colour. The red colour indicates that nitrate is still present and was not reduced previously. An absence of red colour after the addition of zinc dust indicates that no nitrate is present, and thus the nitrate was reduced further than nitrite. Therefore, the nitrate reduction test is evidenced by either the presence of a catabolic end product or the absence of nitrate in the medium.

COMPOSITION

Ingredients	Gms / Ltr
Veg hydrolysate	20.000
Disodium phosphate	2.000
Dextrose (Glucose)	1.000
Potassium nitrate	1.000
Agar	1.000

PRINCIPLE

Due to its nutritive content, the medium supports the growth of aerobes, microaerophiles, facultative and obligate anaerobes. The medium has low agar content which offers varying degree of anaerobiosis and enables organisms with various oxygen requirements to grow. Certain bacteria decomposes amino acid tryptophan from the protein source (Veg hydrolysate) to indole which accumulates in the medium

INSTRUCTION FOR USE

- Dissolve 25.0 grams in 1000 ml purified/distilled water.
- Heat to boiling to dissolve the medium completely.
- Dispense in test tubes.
- Sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes. Cool to 45-50ºC.

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder : Light yellow coloured, may have slightly greenish tinge, homogeneous, free flowing powder.

Appearance of prepared medium : Light amber coloured, clear to slightly opalescent gel forms in tubes as butts.

pH (at 25°C) : 7.2 ± 0.2

INTERPRETATION

Cultural characteristics observed after incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Indole production	Nitrate reduction	Incubation Temperature	Incubation Period
Clostridium perfringens	12924	50-100	Luxuriant	Negative reaction	Positive reaction,red colour developed within 1-2 minutes	35-37°C	18-48 Hours
Clostridium sordellii	9714	50-100	Luxuriant	Positive reaction,red ring at the interface of the medium	Negative reaction	35-37°C	18-48 Hours
Clostridium sporogenes	11437	50-100	Luxuriant	Negative reaction	Negative reaction	35-37°C	18-48 Hours
Escherichia coli	25922	50-100	Luxuriant	Not applicable	Positive reaction,red colour developed within 1-2 minutes	35-37°C	18-48 Hours
Staphylococcus aureus subsp. aureus	25923	50-100	Luxuriant	Negative reaction	Positive reaction,red colour developed within 1-2 minutes	35-37°C	18-48 Hours

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.

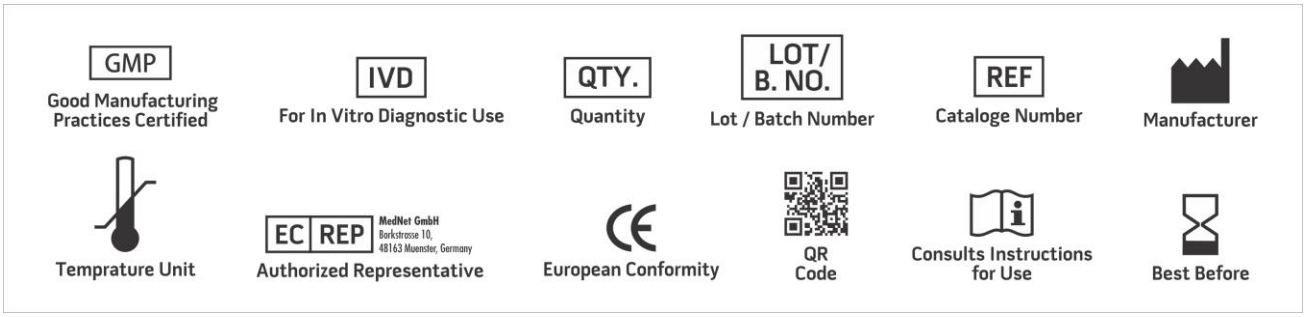
Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

DISPOSAL

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

REFERENCES

1. MacFaddin J.F., 2000(ed), Biochemical Tests for Identification of Medical Bacteria, 3rd edition, Lippincott Williams and Wilkins, New York.
2. Murray PR, Baron, Pfaller, and Yolken (Eds.), 2003, In Manual of Clinical Microbiology, 8th ed., ASM, Washington, D.C.
3. Smith, Rogers and Bettge, 1972, Appl. Microbiol., 23:423.



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

***For Lab Use Only**

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