

TM 2416 - VIBRIO VULNIFICUS AGAR (VVA)

INTENDED USE

For identification of *Vibrio* in accordance with FDA BAM, 1998.

PRODUCT SUMMARY AND EXPLANATION

V. vulnificus has been reported to be an important cause of death due to seafood consumption or after wound infections originating from marine environment. *Vibrio* species in general are alkalophilic and grow well in the presence of relatively high levels of bile salts. This necessitates the use of formulations with alkaline pH for their isolation and identification. Different methods used for the confirmation of *Vibrio* species include physical, biochemical and serological assays. *V. vulnificus* resembles *V. parahaemolyticus* on TCBS agar, but can be differentiated by several biochemical reactions, including beta-galactosidase activity. Identification using oligo nucleotides have also been recommended for the specific identification of the species. The oligonucleotide scheme includes both MPN and direct plating methods followed by hybridization with DNA probes for colony identification. CPC Agar Base, Alkaline peptone water and TCBS Agar are the most used formulations for the isolation of *Vibrios*.

COMPOSITION

| Ingredients | Gms / Ltr |
|------------------|-----------|
| Peptone | 20.000 |
| Sodium Chloride | 30.000 |
| Cellobiose | 10.000 |
| Bromothymol blue | 0.060 |
| Agar | 25.000 |

PRINCIPLE

Vibrio vulnificus Agar is used for the identification of *Vibrio vulnificus* from food samples through oligonucleotide analysis in accordance with FDA BAM, 1998. Peptone provides necessary nitrogenous compounds to the medium. Cellobiose acts as the fermentable carbon source. Sodium chloride maintains the osmotic equilibrium of the medium. Bromothymol blue acts as the indicator dye and agar as the solidifying agent.

INSTRUCTION FOR USE

- Dissolve 85.06 grams in 1000 ml purified / distilled water.
- Heat to boiling to dissolve the medium completely.
- Sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes.
- Cool to 45-50°C. Mix well and pour into sterile Petri plates.

QUALITY CONTROL SPECIFICATIONS

| | |
|-------------------------------|--|
| Appearance of Powder | : Cream to light green homogeneous free flowing powder. |
| Appearance of prepared medium | : Green coloured clear to slightly opalescent gel forms in Petri plates. |
| pH (at 25°C) | : 8.20±0.2 |

INTERPRETATION

Cultural characteristics observed after an incubation.

| Microorganism | ATCC | Inoculum (CFU/ml) | Growth | Recovery | Color of the colony | Incubation Temperature | Incubation Period |
|--------------------------|-------|-------------------|-----------|----------|---|------------------------|-------------------|
| <i>Vibrio vulnificus</i> | 29306 | 50-100 | Luxuriant | >=70% | Yellow opaque colonies (fried egg appearance) | 35-37°C | 18-24 Hours |

PACKAGING:

In pack size of 100 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.

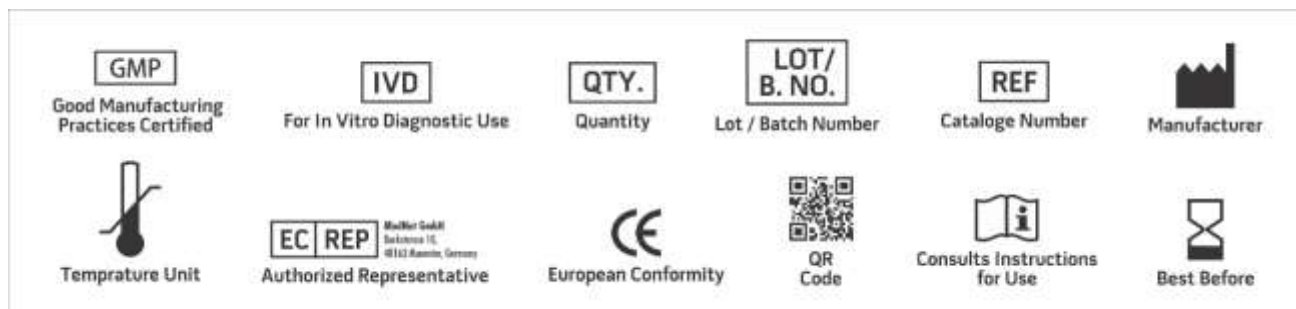
Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

DISPOSAL

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

REFERENCES

- American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D.C.
- FDA, U.S. 1998. Bacteriological Analytical Manual. 8 ed. Gaithersburg, MD: AOAC International.
- Hill, W. E., Keasler, S.P., Trucksess, M.W., Feng, P., Kaysner, C.A. and Lampel, K.A. 1991. Appl. Environ. Microbiol, 57: 707-711.
- Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
- Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
- McPherson, V. L., Watts, J. A., Simpson, L. M. and Oliver, J. D. 1991. Microbios, 67: 141-149.
- Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
- Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.



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

***For Lab Use Only**
Revision: 08 Nov., 2019