

TM 1352 – HAEMOPHILUS TEST AGAR BASE

INTENDED USE

For the susceptibility testing of *Haemophilus influenza*.

PRODUCT SUMMARY AND EXPLANATION

Haemophilus species are nutritionally fastidious in nature. They require either exogenous hemin (X-factor) or NAD (V factor) or both. Due to this reason, Mueller Hinton Agar, which is used for antimicrobial susceptibility of bacteria, can't be used for the antimicrobial susceptibility testing of Haemophilus. Also, addition of blood to Mueller Hinton Agar to supply the essential growth nutrients makes the medium opaque, rendering it unsuitable for antimicrobial susceptibility testing.

Haemophilus Test Agar Base, studied by Jorgensen et al is used for the susceptibility testing of Haemophilus influenzae. This medium has similar composition as Mueller Hinton Agar, with the addition of yeast extract and added growth supplements. Haemophilus Test Agar Base is simple, transparent and possess minimum risk of antagonism of antimicrobial agents. Haemophilus Test Agar Base is also recommended by the United States National Committee for Clinical Laboratory Standards (NCCLS) for both dilution and disc diffusion assays. This medium scores over Mueller Hinton Agar with hemoglobin over clarity, thereby enabling proper visualization of inhibition zones. It also has low levels of the nucleotide thymidine, which allows testing of trimethoprim / sulphamethoxazole.

COMPOSITION

| Ingredients | Gms / Ltr | |
|-------------------------|-----------|--|
| Beef infusion from | 300.000 | |
| Casein acid hydrolysate | 17.500 | |
| Yeast Extract | 5.000 | |
| Starch | 1.500 | |
| Agar | 17.000 | |

PRINCIPLE

Haemophilus Test Agar Base contains beef infusion and casein acid hydrolysate, which provide essential nutrients to the organisms. Yeast extract serves as a source of B complex vitamins. Starch acts as a protective colloid against toxic substances present in the medium. The surface of a Haemophilus Test Agar Base with added nutrients is inoculated either by using swab or by spreading the suspension. Antimicrobial discs i.e. paper discs impregnated with specific amount of antibiotics or other antimicrobial agents are placed on the surface of medium spaced properly. The plates are incubated in a CO2 incubator and subsequently the inhibition zones around each disc are read. Comparing the zones of inhibition with the NCCLS standards, the determination as to whether the organism is susceptible, resistant or intermediate in its response to the antimicrobial substances is made.

INSTRUCTION FOR USE

- Dissolve 21.5 grams in 500 ml distilled water.
- Heat to boiling to dissolve the medium completely.
- Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.
- Cool to 50°C and aseptically add the rehydrated contents of 1 vial of Haemophilus Growth Supplement.
- Mix well and pour into sterile Petri plates.













QUALITY CONTROL SPECIFICATIONS

: Cream to yellow homogeneous free flowing powder Appearance of Powder

: Light amber coloured, clear to slightly opalescent gel forms in Petri plates. Appearance of prepared medium

 $: 7.4 \pm 0.2$ pH (at 25°C)

INTERPRETATION

Cultural characteristics observed with added Haemophilus Growth Supplement in 5-7% carbon dioxide after incubation.

| Microorganism | АТСС | Inoculum (CFU/ml) | Growth | Recovery | Incubation Temperature | Incubation Period |
|---------------------------|-------|----------------------|--------------------|----------|---------------------------|-------------------|
| Haemophilus influenzae | 49766 | 50-100 | Luxuriant | >=70% | 35-37°C | 18 - 24 Hours |
| Enterococcus faecalis | 29212 | 50-100 | Good- luxuriant | >=50% | 35-37°C | 18 - 24 Hours |
| Streptococcus pyogenes | 19615 | 50-100 | Good- luxuriant | >=50% | 35-37°C | 18 - 24 Hours |
| Neisseria meningitidis | 13090 | 50-100 | Good- luxuriant | >=50% | 35-37°C | 18 - 24 Hours |
| Staphylococcus aureus | 25923 | 50-100 | Good- luxuriant | >=50% | 35-37°C | 18 - 24 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



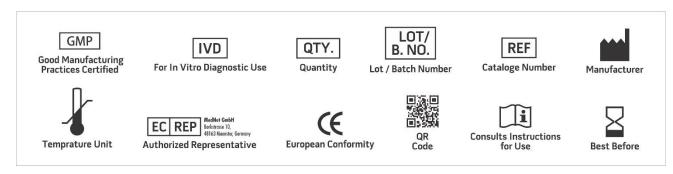








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NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices. *For Lab Use Only

Revision: 08 Nov., 2019







