

# TM 2012 – BORDET GENGOU BROTH

#### **INTENDED USE**

For the selective enrichment of Bordetella pertussis and Bordetella parapertussis.

#### PRODUCT SUMMARY AND EXPLANATION

Bordet Gengou Agar Media were originally formulated by Bordet and Gengou for cultivation of Bordetella species. Bordetella pertussis is the causative agent of whooping cough and with the help of cough-plate technique, B. pertussis can be isolated from pharyngeal extracts, nasopharyngeal secretions and pre-nasal swabs. Kendrick and Eldering modified the original media by replacing 50% human or rabbit blood with 15% sheep blood to make the medium more enriched for detection of B.pertussis by the virtue of its haemolytic reaction. Enrichment of the basal media with 25% human blood aids in the detection of Mycobacterium species from small sputum inocula and in Streptomycin sensitivity testing. The medium is highly nutritious thus supports luxuriant growth of Bordetella species and can also be used for mass cultivation of B. pertussis for vaccine production and for maintaining stock cultures. Bordetella Broth Base is prepared with the omision of Agar and is recommended for the selective enrichment of Bordetella species.

#### **COMPOSITION**

Ingredients	Gms / Ltr
Potatoes, infusion from	125.000
Peptone	10.000
Sodium chloride	5.500

### **PRINCIPLE**

Potato infusion and peptone serve as carbon and nitrogen source, long chain amino acids, essential nutrients while glycerol and blood enrichment provides additional nutrients. Sodium chloride maintains osmotic equilibrium. Incubation should be carried out in a moist chamber (60% humidity) at 37°C for upto 7 days. Medium should not be over dried before use. After 40 hours B.pertussis colonies appear smooth, raised, glistening with a zone of haemolysis. Some strains of Bordetella are not haemolytic. For confirmation, serodiagnosis and biochemical test should be performed. This medium can be made more selective for Bordetella, by using antibiotics like penicillin, methicillin, cephalexin of which, cephalexin was found to be superior. Cephalexin suppresses unwanted nasopharyngeal growth and significantly increases the isolation rate of Bordetella species. Cephalexin is used at a concentration of 40 mg/liter (TS 012). Amphotericin B (10 µg/ ml) can be added as an antifungal agent to the medium. For isolation of I from specimens, use standard procedures. Incubate the plates in a moist chamber at 35-37°C for 7 days and examine daily with or without dissecting microscope (oblique illumination) to detect the presence of *B. pertussis*.

# **INSTRUCTION FOR USE**

- Dissolve 20.00 grams in 1000 ml distilled water containing 10 ml glycerol.
- Heat if necessary to dissolve the medium completely. Sterilize by autoclaving at 15 psi (121°C) for 15 minutes.
- Cool to 45-50°C and aseptically add 15-20% sterile, fresh defibrinated blood (sheep, rabbit, human or horse).
- For selectivity aseptically add rehydrated contents of 2 vials of Bordetella Selective supplement (TS 012).
- Mix thoroughly and distribute into sterile tubes or flasks as desired.

### **QUALITY CONTROL SPECIFICATIONS**













**Appearance of Powder** : Cream to yellow homogeneous free flowing powder

: Basal Medium: Light yellow coloured clear to slightly opalescent solution. After Appearance of prepared medium

addition of glycerol and 15% v/v sterile defibrinated blood: Cherry red

coloured opaque solution.

: 6.7±0.2 pH (at 25°C)

### **INTERPRETATION**

Cultural characteristics observed after incubation.

Microorganism	АТСС	Growth	Inoculum	Incubation Temperature	Incubation Period
Staphylococcus aureus subsp.aureus	25923	Inhibited	>=10 <sup>3</sup>	35-37°C	3-4 Days
Bordetella bronchiseptica	4617	Good-luxuriant	50-100	35-37°C	3-4 Days

#### **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.

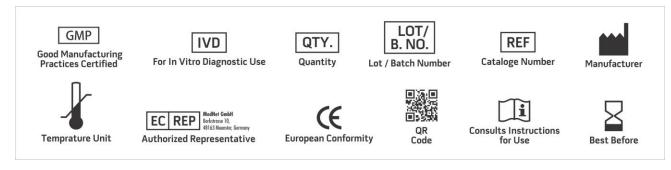
Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

### **DISPOSAL**

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

# **REFERENCES**

- 1. Kendrick and Eldering, 1934, Am. J. Public Health, 24:309
- 2. Suitcliffe E. M. and Abbott J. D., 1972, B. M. J., iii:732.
- 3. Bordet and Gengou, 1906, Ann. Inst. Pasteur, 20:731.
- 4. Broome C. V., Fraser D. W. and English J. W., 1979, Internat. Symp. on Pertussis, DHEW, Washington, D.C., 19.
- 5. Flemming A., 1932, J. Path. Bacteriol., 35:831.

















**NOTE:** Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices. \*For Lab Use Only Revision: 08 Nov., 2019









