# BORDET GENGOU AGAR

## INTENDED USE

Remel Bordet Gengou Agar, when enriched with blood, is a solid medium recommended for use in qualitative procedures for the isolation of *Bordetella pertussis* and *Bordetella parapertussis*.

## SUMMARY AND EXPLANATION

Whooping cough is caused by *B. pertussis* and *B. parapertussis* which are among the most fastidious bacteria known and require special culturing techniques. This medium was originally described by Bordet and Gengou in 1906 and included 50% blood.<sup>1</sup> Kendrick and Eldering reduced the amount of animal blood to 15% making Bordet Gengou medium more practical for use in the clinical laboratory.<sup>2</sup> In separate studies, Ahmad and Kurzynski reported maximum sensitivity is obtained by routinely using both Bordet Gengou Agar and a selective charcoal blood agar for primary isolation of *Bordetella* spp.<sup>3.4</sup> The CDC Pertussis Laboratory recommends including a nonselective medium, such as Bordet Gengou, along with selective media to increase the recovery of *Bordetella* spp.<sup>5</sup> Bordet Gengou Agar has the added advantage of being able to detect hemolysis by *B. pertussis*.

#### PRINCIPLE

Bordet Gengou Agar is an enriched casein peptone medium with potato infusion and glycerol to supply nutrients which support the growth of *Bordetella* spp. Blood is added to provide additional nutrients and to enable the detection of hemolytic reactions. Sodium chloride maintains osmotic equilibrium.

## **REAGENTS (CLASSICAL FORMULAE)\***

Potato Infusion	g
Sodium Chloride5.0	g
Meat Peptone	g
Casein Peptone	g

Glycerol1	0.0	ml
Sheep Blood	. 15	%
Agar1	5.0	g
Demineralized Water	0.0	mĪ

pH 7.4 ± 0.2 @ 25°C

\*Adjusted as required to meet performance standards.

## PROCEDURE

All suspected cases of pertussis should have a nasopharyngeal (NP) aspirate and/or NP swab obtained for culture. Specimens should be collected from the posterior nasopharynx up to 4 weeks after onset of symptoms. NP aspirates are the preferred method of specimen collection because they have a higher recovery rate of *Bordetella* spp. than NP swabs. If swabs are used, only calcium alginate or polyester swabs are acceptable. Cotton swabs may contain inhibitors that decrease isolation rates. Consult appropriate references for detailed specimen collection guidelines.<sup>5-7</sup>

Optimally, media should be inoculated directly at the time of specimen collection. If a delay cannot be avoided, the specimen should be placed in a transport system, such as Regan-Lowe Semisolid Transport Medium (REF R064141). The CDC Pertussis Laboratory recommends shipping clinical specimens for recovery of *Bordetella* spp. at 4°C. Incubation of transport medium prior to shipment to the laboratory has been shown to result in overgrowth of other flora and decreased yield of *B. pertussis.*<sup>6</sup>

- 1. Inoculate specimen onto Bordet Gengou Agar as soon as possible after receipt in the laboratory and streak for isolation. Both selective and nonselective media should be inoculated.
- 2. Incubate plates in ambient air at 35-36°C for at least 7 days; incubation up to 12 days may increase yield.<sup>5</sup>

**Note:** *B. pertussis* will not grow at 37°C; incubation at 35°C is optimal.<sup>5</sup> The CDC Pertussis Laboratory recommends incubation at 35-36°C.<sup>6</sup>

- 3. To avoid drying, place plates in a plastic bag or moist chamber with a sterile moistened filter paper; drying reduces the recovery rate of *B. pertussis*.<sup>6</sup>
- 4. If specimen is received in Regan-Lowe Semisolid Transport Medium, it should be incubated along with the plates and subcultured after 48 hours to selective and nonselective media.<sup>7</sup>
- 5. Examine plates daily for growth. Bordetella colonies are tiny, smooth, transparent, glistening, and domed.

**Pour Tube:** Melt the agar in a boiling water bath and cool to 45-50°C. Aseptically add 3 ml of sterile sheep, rabbit, or horse blood to the tube. Mix and dispense into a sterile petri dish.

# QUALITY CONTROL

All lot numbers of Bordet Gengou Agar have been tested using the following quality control organisms and have been found to be acceptable. This quality control testing conforms with or exceeds CLSI standards.<sup>8</sup> Testing of control organisms should be performed in accordance with established laboratory quality control procedures. If aberrant quality control results are noted, patient results should not be reported.

#### CONTROL

Bordetella pertussis ATCC<sup>®</sup> 12742 Streptococcus sanguis ATCC<sup>®</sup> 10556 INCUBATION Aerobic, up to 5 days @ 35-36°C Aerobic, 18-24 h @ 35-36°C RESULTS Growth Growth

# LIMITATIONS

Specimens collected prior to the administration of antibiotics have higher recovery rates for Bordetella.<sup>5,7</sup> 1.

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Refer to the front of Remel Technical Manual of Microbiological Media for General Information regarding precautions, product storage and deterioration, specimen collection, storage and transportation, materials required, quality control, and limitations.

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