



QUALITY CONTROL PROCEDURES

I INTRODUCTION

Bordet Gengou Agar Base, with the addition of glycerol and sterile blood, is used in qualitative procedures for the detection and isolation of *Bordetella pertussis* from clinical specimens.

II PERFORMANCE TEST PROCEDURE

1. Inoculate representative samples with the cultures listed below.
 - a. Streak inoculate the plates with 10³ – 10⁴ CFU using a 10 µL (0.01 mL) loop.
 - b. Incubate at 35 ± 2 °C in an aerobic atmosphere supplemented with 3-5% carbon dioxide.
2. Examine plates for up to 7 days for growth and zones of hemolytic activity.
3. Expected Results

Organisms	ATCC®	Recovery	Hemolysis
* <i>Bordetella pertussis</i>	9797	Moderate to heavy growth	Beta
<i>Staphylococcus aureus</i>	25923	Fair to heavy growth	None
* <i>Streptococcus pneumoniae</i>	6305	Fair to heavy growth	Alpha

*Recommended organism strain for User Quality Control.

III ADDITIONAL QUALITY CONTROL

1. Examine plates as described under "Product Deterioration."
2. Visually examine representative plates to assure that any existing physical defects will not interfere with use.
3. Note the firmness of plates during the inoculation procedure.
4. Incubate uninoculated representative plates at 33 – 37 °C for 72 h and examine for microbial contamination.

PRODUCT INFORMATION

IV INTENDED USE

This medium is used in qualitative procedures for detection and isolation of *Bordetella pertussis* from clinical specimens.

V SUMMARY AND EXPLANATION

Bordet Gengou medium is used in clinical laboratories as a method of diagnosing whooping cough. *Bordetella pertussis*, the etiologic agent of this disease, may be isolated on cough plates, from nasopharyngeal swabs and other sources of pharyngeal exudate.

Bordet and Gengou introduced the medium in 1906 as a method of maintaining stock cultures.¹ In 1934, Kendrick and Eldering replaced the 50% human or rabbit blood recommended in the original formulation with 15% sheep blood to make the medium more practical for laboratories to produce for routine clinical procedures.²

VI PRINCIPLES OF THE PROCEDURE

Bordet Gengou Blood Agar is an enriched, casein peptone medium with potato infusion and glycerol to supply the nutrients necessary to support the growth of *B. pertussis*. Defibrinated sheep blood supplies additional nutrients and enables the detection of hemolytic reactions, which aid in the identification of *B. pertussis*.

VII REAGENTS

Bordet Gengou Blood Agar

Approximate Formula* Per Liter Purified Water

Potato, Infusion from (solids).....	4.5 g	Agar	20.0 g
Pancreatic Digest of Casein	5.0 g	Glycerol	10.0 mL
Peptic Digest of Animal Tissue	5.0 g	Sheep Blood, defibrinated	15%
Sodium Chloride	5.5 g		

*Adjusted and/or supplemented as required to meet performance criteria.

Warnings and Precautions: For *in vitro* Diagnostic Use.

If excessive moisture is observed, invert the bottom over an off-set lid and allow to air dry in order to prevent formation of a seal between the top and bottom of the plate during incubation.

Pathogenic microorganisms, including hepatitis viruses and Human Immunodeficiency Virus, may be present in clinical specimens. "Standard Precautions"³⁻⁶ and institutional guidelines should be followed in handling all items contaminated with blood and other body fluids. After use, prepared plates, specimen containers and other contaminated materials must be sterilized by autoclaving before discarding.

Storage Instructions: On receipt, store plates in the dark at 2 – 8 °C. Avoid freezing and overheating. Do not open until ready to use. Minimize exposure to light. Prepared plates stored in their original sleeve wrapping at 2 – 8 °C until just prior to use may be inoculated up to the expiration date and incubated for recommended incubation times. Allow the medium to warm to room temperature before inoculation.

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

VIII SPECIMEN COLLECTION AND HANDLING

Refer to appropriate texts for details of specimen collection and handling procedures.⁷⁻¹⁰

Specimens should be obtained before antimicrobial agents have been administered. Provision must be made for prompt delivery to the laboratory.

IX PROCEDURE

Material Provided: Bordet Gengou Blood Agar

Materials Required But Not Provided: Ancillary culture media, reagents, quality control organisms and laboratory equipment as required.

Test Procedure: Observe aseptic techniques.

The agar surface should be smooth and moist, but without excessive moisture.

Inoculate the medium as soon as possible after the specimen arrives at the laboratory. To culture a specimen from a swab, inoculate the medium by rolling the swab over a third of the agar surface, and streak the remainder of the plate to obtain isolated colonies. Material not being cultured from swabs may be streaked onto the medium with a sterile inoculating loop. The streak plate technique is used primarily to obtain isolated colonies from specimens containing mixed flora.

For "cough" plate inoculation, hold the Bordet Gengou Agar plate about 10 cm away from the patient's mouth during an attack of coughing.

Incubate the plates in an inverted position (agar side up) in a moist chamber at 35 °C for 7 days. Colonies of *B. pertussis* may not be visible without the aid of a microscope for 2 – 4 days. Plates may be discarded as negative after 7 days of incubation.

User Quality Control: See "Quality Control Procedures."

Quality control requirements must be performed in accordance with applicable local, state and/or federal regulations or accreditation requirements and your laboratory's standard Quality Control procedures. It is recommended that the user refer to pertinent CLSI guidance and CLIA regulations for appropriate Quality Control practices.

X RESULTS

Examine the plates daily with and without a dissecting microscope (oblique illumination) to detect the presence of *Bordetella pertussis*. To prevent overgrowth by spreading colonies or molds, use a sterile scalpel or needle to remove the portions of the agar that contain these contaminants.

Bordetella pertussis produces small, domed, glistening colonies that resemble bisected pearls. The colonies are usually surrounded by a zone of hemolysis. However, some strains of *B. pertussis* are not hemolytic. Gram staining, biochemical tests and serological procedures should be performed to confirm findings.

XI LIMITATIONS OF THE PROCEDURE

This prepared plated medium is intended for primary isolation. Some diagnostic tests may be performed with the primary plate. However, a pure culture is recommended for biochemical tests and serological procedures. Consult appropriate texts for further information.^{7,9,11}

XII AVAILABILITY

Cat. No.	Description
297876	BD BBL™ Bordet Gengou Blood Agar

XIII REFERENCES

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Technical Information: In the United States, contact BD Technical Service and Support at 800-638-8663 or www.bd.com/ds.



Becton, Dickinson and Company
7 Loveton Circle
Sparks, MD 21152 USA



Benex Limited
Pottery Road, Dun Laoghaire
Co. Dublin, Ireland

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