



BD™ Bordet Gengou Agar with 15% Sheep Blood • BD Bordetella Agar with Charcoal and 7% Horse Blood

INTENDED USE

BD Bordet Gengou Agar with 15% Sheep Blood and **BD Bordetella Agar with Charcoal and 7% Horse Blood** are selective media for the isolation of *Bordetella pertussis* and *B. parapertussis*.

PRINCIPLES AND EXPLANATION OF THE PROCEDURE

Microbiological method.

All *Bordetella* species are respiratory pathogens, residing in ciliated epithelial cells of the respiratory tract. *B. pertussis* and *B. parapertussis* are uniquely human pathogens. *B. pertussis* is the major cause of whooping cough. *B. parapertussis* is associated with a milder form of this disease. *B. bronchiseptica* is primarily an animal pathogen, but may produce bronchitis, pneumonia and extra-respiratory tract infections in humans.

Bordet Gengou Agar Base is a modification of the medium originally described by Bordet and Gengou for the cultivation of *Bordetella pertussis*.¹ **BD Bordet Gengou Agar with 15% Sheep Blood** is prepared according to the modified formula recommended by the American Public Health Association.² In this medium, infusion from potato provides nitrogen and vitamins. Many compounds may inhibit growth of *B. pertussis*, including fatty acids present in nasal secretions or cotton from the collection swab. Starch, present from the potato infusion, absorbs fatty acids. Glycerol is a carbon source. Sodium chloride maintains the osmotic balance of the medium. The addition of 15% sheep blood provides essential complex growth factors.

BD Bordetella Agar with Charcoal and 7% Horse Blood is based on the formula of Mishulow et al. who found it to be an efficient substitute for Bordet-Gengou Agar.³ Later, this medium was further developed and became known as Regan-Lowe Charcoal Agar.⁴ In this medium, beef extract and gelatin peptone provide nutrients. Starch and charcoal act as adsorbents of toxic fatty acids, radicals and peroxides, replacing the potato infusion in Bordet Gengou Agar. Niacin is a vitamin that promotes growth. Horse blood provides complex growth factors and further reduces the toxic effect of peroxides.

In **BD Bordet Gengou Agar with 15% Sheep Blood** and **BD Bordetella Agar with Charcoal and 7% Horse Blood** cefalexin is added as a selective agent to partially suppress normal respiratory tract flora.^{5,6}

REAGENTS

Formulas* Per Liter Purified Water

BD Bordet Gengou Agar with 15% Sheep Blood		BD Bordetella Agar with Charcoal and 7% Horse Blood	
Potato, Infusion from Solids	4.5 g	Beef Extract	10.0 g
Pancreatic Digest of Casein	5.0	Pancreatic Digest of Gelatin	10.0
Peptic Digest of Animal Tissue	5.0	Soluble Starch	10.0
Sodium Chloride	5.5	Charcoal (activated)	4.0
Agar	20.0	Sodium Chloride	5.0
Cefalexin	0.04	Niacin (Nicotinic acid)	0.01
Glycerol	10.0 ml	Cefalexin	0.04
Sheep Blood, defibrinated	15%	Horse Blood, defibrinated	7%
pH 7.2 +/- 0.2		Agar	12.0 g
		pH 7.4 +/- 0.2	

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

PRECAUTIONS

IVD . For professional use only.

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

Consult **GENERAL INSTRUCTIONS FOR USE** document for aseptic handling procedures, biohazards, and disposal of used product.

STORAGE AND SHELF LIFE

On receipt, store plates in the dark at 2 to 8° C, in their original sleeve wrapping until just prior to use. Avoid freezing and overheating. The plates may be inoculated up to the expiration date (see package label) and incubated for the recommended incubation times.

Plates from opened stacks of 10 plates can be used for one week when stored in a clean area at 2 to 8° C.

USER QUALITY CONTROL

Inoculate representative samples with the following strains (for details, see **GENERAL INSTRUCTIONS FOR USE** document). Incubate aerobically in a moist chamber for 5 to 7 days at 35 to 37° C. Do not incubate in an aerobic atmosphere enriched with carbon dioxide.

Strains	BD Bordet Gengou Agar with 15% Sheep Blood	BD Bordetella Agar + Charcoal + 7 % Horse Blood
<i>Bordetella pertussis</i> ATCC™ 9797	Growth fair to excellent	Growth good to excellent
<i>Bordetella parapertussis</i> ATCC 15311	Growth fair to excellent	Growth good to excellent
<i>Streptococcus pneumoniae</i> ATCC 6305	Inhibition partial to complete	Inhibition partial to complete
<i>Streptococcus pyogenes</i> ATCC 19615	Inhibition partial to complete	Inhibition partial to complete
Uninoculated	Dark red with brownish hue, opaque	Shiny black with a reddish hue, opaque

PROCEDURE

Materials Provided

BD Bordet Gengou Agar with 15% Sheep Blood or **BD Bordetella Agar with Charcoal and 7% Horse Blood**, both provided in 90 mm **Stacker™** plates. Microbiologically controlled.

Materials Not Provided

Ancillary culture media, reagents and laboratory equipment as required.

Specimen Collection and Specimen Types

The “cough plate” method for the diagnosis of whooping cough is no longer recommended.⁷ Secretions from the posterior nasopharynx, nasopharyngeal lavage, or a nasopharyngeal swab (calcium alginate on a wire handle) should be collected within the first week of paroxysmal coughing.³ Do not use cotton swabs! The collection of secretions by vacuum suction is recommended. Specimens from the throat should not be used. Special transport media, based on charcoal, have been described. It is mandatory that the specimens are transported to the laboratory as fast as possible, even if transport media are used. Transport times of > 24 hours will reduce the viability of *Bordetella* significantly. For details, consult the references.^{6,8,9}

Test Procedure

Inoculate the plates as soon as possible after the arrival of the specimens in the laboratory and streak for dilution. Incubate the plates in ambient air in a moist chamber (e.g., a **BD GasPak™** jar containing some water in a beaker) for 7 to 10 days, and inspect daily, using a magnifying lens. Do not incubate in an aerobic atmosphere enriched with carbon dioxide since the gas

may suppress the organism. The optimal incubation temperature is 34 to 36° C. Higher temperature may reduce the growth of *Bordetella*.⁶

Results

After sufficient incubation, *B. pertussis* colonies on **BD Bordet Gengou Agar with 15% Sheep Blood** and on **BD Bordetella Agar with Charcoal and 7% Horse Blood** will be small, white, domed and glistening, resembling bisected pearls. On **BD Bordet Gengou Agar with 15% Sheep Blood**, colonies of *B. pertussis* and *B. parapertussis* may be surrounded by a weak beta-hemolysis.

PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE

BD Bordet Gengou Agar with 15% Sheep Blood and **BD Bordetella Agar with Charcoal and 7% Horse Blood** are approved selective isolation media for *Bordetella* species, including *B. pertussis* and *B. parapertussis*.^{6,9} Most recent data support the use of the charcoal based medium^{6,10,11}

Cefalexin is included into these media as an inhibitor of many Gram positive and certain Gram negative bacteria present in the normal throat flora, but is not completely inhibitory to all organisms.^{5,6}

Bordetella bronchiseptica may grow on these media. It is an opportunistic pathogen which may also produce respiratory infections.¹²

Further tests are necessary for the complete identification of *Bordetella pertussis* and *B. parapertussis*. The direct fluorescent antibody (DFA) test which can also be used along with culture for the direct detection of the agents from the specimens, can also be used for confirmation of the isolates obtained on these media.^{6,8,9}

REFERENCES

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PACKAGING/AVAILABILITY

BD Bordet Gengou Agar with 15% Sheep Blood

Cat. No. 254400

Ready-to-use Plated Media, cpu 20

BD Bordetella Agar with Charcoal and 7 % Horse Blood
Cat. No. 256054 Ready-to-use Plated Media, cpu 20

FURTHER INFORMATION

For further information please contact your local BD representative.



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