BD™ Gardnerella Selective Agar with 5% Human Blood

INTENDED USE

BD Gardnerella Selective Agar with 5% Human Blood is a partially selective and differential medium for the isolation of *Gardnerella vaginalis* from clinical specimens.

PRINCIPLES AND EXPLANATION OF THE PROCEDURE

Microbiological method.

Gardnerella vaginalis is considered to be one of the organisms causing vaginitis.[1-4] Although the organism may be present in a high percentage of normal women in the vaginal flora, its importance as a cause of non-specific vaginitis (also called bacterial vaginosis) has never been questioned. In symptomatic women, *G. vaginalis* frequently is associated with anaerobes such as *Prevotella bivia*, *P. disiens*, *Mobiluncus*, *Peptostreptococcus,* and/or others which are a regular part of the urethral or intestinal, but not vaginal flora. In non-specific vaginitis, normal *Lactobacillus* flora is reduced or absent. *Gardnerella vaginalis* is considered to be the indicator organism for non-specific vaginitis which, in fact, is a polymicrobial infection.[3,4] Although non-culture methods such as a direct Gram stain have been recommended in recent years for genital specimens, culture is still preferred by many laboratories.[1,5] *G. vaginalis* may also be responsible for a variety of other diseases such as preterm birth, chorioamnionitis, urinary tract infections, newborn infections, and septicemia.[6]

The detection of the organism on routinely used media is difficult since *Gardnerella* and other bacteria such as lactobacilli and streptococci may produce alpha hemolysis on sheep blood containing media. On media containing human blood, however, *Gardnerella vaginalis* produces a characteristic beta-hemolysis.[1,7-9] Since *Gardnerella vaginalis*, although often considered Gram negative, is a Gram positive bacterium, it may be selected on media containing colistin and nalidixic acid.[1,10,11]

BD Gardnerella Selective Agar with 5% Human Blood is based on Columbia CNA Agar which provides nutrients and the inhibitors colistin and nalidixic acid which inhibit Gram negative but not Gram positive bacteria. The medium is supplemented with Proteose Peptone to improve the growth of *Gardnerella*. Amphotericin B is added to reduce the growth of yeasts (e.g. *Candida*) which are also frequently present in vaginal specimens. Human blood is added as a nutrient and to detect the characteristic diffuse beta hemolysis of the organism.

REAGENTS

BD Gardnerella Selective Agar with 5% Human Blood

Formula* Per Liter Purified Water

Pancreatic Digest of Casein	12.0 g
Peptic Digest of Animal Tissue	5.0
Proteose Peptone	10.0
Yeast Extract	3.0
Beef Extract	3.0
Corn Starch	1.0
Sodium Chloride	5.0
Agar	13.5
Colistin	0.01
Nalidixic Acid	0.01
Amphotericin B	0.004
Human Blood, defibrinated	5%

pH 7.3 +/- 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.

WARNING:

Contains potentially biohazardous material. The human blood used for the preparation of this medium has been tested for the presence of HIV, HBsAg and other types of hepatitis using the criteria and methods presently used by blood banks to screen human blood used for transfusion and was found to be nonreactive. Because no test method currently available can offer complete assurance that HIV, hepatitis and other infectious agents are absent, *specimens and this medium should be handled as though capable of transmitting an infectious disease.* Therefore, it is recommended to handle this medium according to Biosafety Level 2.

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 in a carbon dioxide enriched atmosphere for 48 to 72 hours at 35 to 37° C. Alternatively, the inoculated medium may be incubated in a microaerobic atmosphere, e.g., by using the **BD CampyPak™** system.

Strains	Test Results
Gardnerella vaginalis ATCC™ 14018	Growth good to excellent; small grey white colonies surrounded by a diffuse beta-hemolysis
Escherichia coli ATCC 25922	Inhibition complete
Proteus mirabilis ATCC 14153	Inhibition partial to complete; no swarming
Candida albicans ATCC 10231	Inhibition partial to complete
Uninoculated	Red (blood color)

PROCEDURE

Materials Provided

BD Gardnerella Selective Agar with 5% Human Blood (90 mm **Stacker™** plates). Microbiologically controlled.

Materials Not Provided

Ancillary culture media, reagents and laboratory equipment as required.

Specimen Types

Vaginal swabs or other specimens suspected to contain *G. vaginalis* are suitable. Ideally, two swabs should be collected, one for the culture and one for a direct Gram stain. A transport medium (e.g., **BD Port-A-Cul™**) must be used if the specimens are not immediately processed. (see also **PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE**).

Test Procedure
Streak the specimens onto **BD Gardnerella Selective Agar with 5% Human Blood** by an approved dilution technique. Also inoculate **BD Columbia Agar with 5% Sheep Blood**. Incubate both plates in a CO_2 enriched aerobic atmosphere at 36 +/- 2° C for 48 to 72 hours. Also, a direct Gram stain should be prepared from all vaginal specimens.[1,2,5]

Results
After the incubation, inspect **BD Gardnerella Selective Agar with 5% Human Blood** for the presence of small to medium colonies, surrounded by a diffuse beta-hemolysis. Compare this result with the growth obtained on the **BD Columbia Agar with 5% Sheep Blood** plate. If beta hemolytic colonies appear only on the selective *Gardnerella* medium, the presence of *G. vaginalis* is highly probable. If beta hemolytic colonies appear on both media, make sure by a Gram stain that the organism on the selective *Gardnerella* medium is a Gram variable, small, diphtheroid rod. The presence of cocci in the Gram stain from a beta hemolytic colony on **BD Gardnerella Selective Agar with 5% Human Blood** indicates that the isolated organisms are beta hemolytic streptococci or staphylococci, but not *G. vaginalis*. Alpha and non-hemolytic colonies can be disregarded. On media containing sheep blood, *Gardnerella* produces small alpha- or non-hemolytic colonies. In case of a mixed culture with other beta-hemolytic organisms, eventually subculture isolated colonies from **BD Gardnerella Selective Agar with 5% Human Blood** again on both media and incubate as indicated above. Further biochemical tests are necessary for the final identification of the isolates. Consult the references.[1,7,9]

In the direct Gram stain of vaginal specimens, “clue cells” (= vaginal epithelial cells loaded with masses of short, Gram variable rods) will be visible if the patient suffers from bacterial vaginosis.[1,5]

Interpretation of the Results
If *G. vaginalis* has been isolated from vaginal specimens, the resulting diagnosis must be carefully compared with the subjective and objective clinical symptoms. Typically, if *Gardnerella* is the cause of bacterial vaginosis, the patients complain of an increased, malodorous discharge. The pH of the secretions is >4.5, and “clue cells” are present in the Gram stain. For details, consult the references.[1-3,5]

PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE
BD Gardnerella Selective Agar with 5% Human Blood is a partially selective medium for the isolation of *Gardnerella vaginalis*.[4,11] *G. vaginalis* will produce beta-hemolytic colonies on the medium with human blood, and alpha-hemolytic colonies on media containing sheep blood.
BD Gardnerella Selective Agar with 5% Human Blood is not inhibitory to other Gram positive organisms such as streptococci, staphylococci, or *Listeria* which may or may not produce beta hemolysis on media containing human blood and may also occur in vaginal specimens. Therefore, microscopic and biochemical tests are required to confirm the identity of an isolate as *G. vaginalis*.
The presence of *G. vaginalis* in a vaginal specimen does not necessarily indicate that the isolated organism is the cause of an infection.
Do not use **BD Gardnerella Selective Agar with 5% Human Blood** for the determination of hemolytic reactions of organisms other than *Gardnerella vaginalis*.

REFERENCES

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PACKAGING/AVAILABILITY

BD Gardnerella Selective Agar with 5% Human Blood

Cat. No. 254094 Ready-to-use Plated Media, cpu 20
Cat. No. 257664 Ready-to-use Plated Media, cpu 120

FURTHER INFORMATION

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