



BBL™ Rapid Fermentation Medium, Base
BBL™ Rapid Fermentation Medium, Dextrose
BBL™ Rapid Fermentation Medium, Lactose
BBL™ Rapid Fermentation Medium, Maltose
BBL™ Rapid Fermentation Medium, Sucrose



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QUALITY CONTROL PROCEDURES

I INTRODUCTION

Rapid Fermentation Medium is used for the differentiation of *Neisseria* species based upon their carbohydrate fermentation patterns.

II PERFORMANCE TEST PROCEDURE

1. Inoculate representative samples with the cultures listed below.
 - a. Inoculate the tubes by depositing a loopful (0.01 mL calibrated loop) of inoculum below the surface of the medium and mix well. Use 18- to 24-h growth from either Chocolate II Agar plates or Chocolate II Agar slants as inocula.
 - b. Incubate tubes with loosened caps at 35 ± 2°C in an aerobic atmosphere without supplementation with carbon dioxide.
2. Examine tubes periodically after 4 h for acid production (yellow color). Continue incubation overnight, if necessary.
3. Expected Results

Production of Acid from:

Organisms	ATCC™	Dextrose	Lactose	Maltose	Sucrose
* <i>Neisseria gonorrhoeae</i>	43070	+	-	-	-
* <i>Neisseria meningitidis</i>	13090	+	-	+	-
* <i>Neisseria lactamica</i>	23970	+	+	+	-
* <i>Neisseria sicca</i>	29193	+	-	+	+

The basal medium is negative with all organisms.

*Recommended organism strain for User Quality Control.

III ADDITIONAL QUALITY CONTROL

1. Examine tubes as described under "Product Deterioration."
2. Visually examine representative tubes to assure that any existing physical defects will not interfere with use.
3. Determine the pH potentiometrically at room temperature for adherence to the specification of 7.3 ± 0.2.
4. Incubate uninoculated representative tubes at 20–25°C and 30–35°C and examine after 7 days for microbial contamination.

PRODUCT INFORMATION

IV INTENDED USE

Rapid Fermentation Medium provides a rapid method for the differentiation of *Neisseria* species isolated from clinical specimens.

V SUMMARY AND EXPLANATION

The occurrence of *Neisseria gonorrhoeae* in extra-genitourinary sites has made it important to differentiate these organisms from other *Neisseria* species.^{1,2} The isolation of *N. meningitidis* and *N. lactamica* from genitourinary sites further indicates the need for differentiation of these species.¹⁻³

The classical method for the differentiation of *Neisseria* species is based upon the determination of their carbohydrate utilization patterns. Pathogenic *Neisseria* are extremely fastidious organisms in both their growth and metabolic activities, thereby requiring an enriched culture medium. The conventional enriched medium for the determination of carbohydrate utilization is Cystine **Trypticase™** Agar medium (**CTA Medium™**) containing 0.5 to 1% carbohydrate.³ The test battery includes dextrose, lactose, maltose, and sucrose and an identification can usually be made in 24 h.

Rapid Fermentation Medium is a modification of the standard **CTA Medium**. The modified formula includes an increased agar and carbohydrate content that, when exposed to a large number of organisms, exhibits an acid shift by the phenol red indicator. An identification often can be made within 4 h.

VI PRINCIPLES OF THE PROCEDURE

Rapid Fermentation Medium is a modification of **CTA Medium** which produces rapid results in detecting acid production from dextrose, lactose, maltose and sucrose. Although much of the literature refers to fermentation patterns for *N. gonorrhoeae*, it has been shown that this species metabolizes dextrose by strictly aerobic mechanisms; i.e., by a combination of the Entner-Doudoroff and pentose phosphate pathways. The utilization of dextrose by *N. gonorrhoeae*, as indicated by an acid change in the pH indicator present in the medium, is due to the production of acetic acid and small amounts of lactic acid.⁴ The negative carbohydrate test results from the deamination of the peptone in the absence of any utilizable carbohydrate.

VII REAGENTS

Rapid Fermentation Medium, Base

Approximate Formula* Per Liter Purified Water

Cystine	0.5 g	Sodium Chloride	5.0 g
Pancreatic Digest of Casein.....	20.0 g	Sodium Sulfite	0.5 g
Agar	3.5 g	Phenol Red	0.017 g

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

Rapid Fermentation Medium with Dextrose, Lactose, Maltose or Sucrose contains the above ingredients with 20 g/L of the appropriate carbohydrate.

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

Tubes with tight caps should be opened carefully to avoid injury due to breakage of glass.

Observe aseptic techniques and established precautions against microbiological hazards throughout all procedures. After use, prepared tubes, specimen containers and other contaminated materials must be sterilized by autoclaving before discarding.

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

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

VIII SPECIMEN COLLECTION AND HANDLING

Specimens suitable for culture may be handled using various techniques. For detailed information, consult appropriate texts.^{5,6} Specimens should be obtained before antimicrobial agents have been administered. Provision must be made for prompt delivery to the laboratory.

IX PROCEDURE

Material Provided: Rapid Fermentation Medium, Base or Rapid Fermentation Medium, Dextrose or Rapid Fermentation Medium, Lactose or Rapid Fermentation Medium, Maltose or Rapid Fermentation Medium, Sucrose

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

Test Procedure: Observe aseptic techniques.

1. Remove a full loopful of fresh colony growth from the surface of a Chocolate II Agar plate or slant. A large inoculum must be used in order to obtain a rapid reaction.
2. Deposit the inoculum below the surface of the medium and mix well.
3. Repeat for each tube to be inoculated.
4. Incubate all tubes at 35 ± 2°C in an aerobic atmosphere without carbon dioxide. Observe periodically after 4 h for reactions noted below. Continue incubation overnight if necessary. A few strains may require incubation for up to 48–72 h.

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 (formerly NCCLS) guidance and CLIA regulations for appropriate Quality Control practices.

X RESULTS

1. After incubation, compare the reactions produced by the unknown isolates with those produced by the known control organisms. Negative reactions are red. A positive reaction is indicated by a change of the phenol red indicator from red to yellow. The control cultures should produce results as shown in the table. If the results with the control cultures do not agree with those in the table, review the procedure, check the control cultures by Gram staining and performing the oxidase test and repeat the fermentation test if necessary.
2. If no positive carbohydrate reactions are observed within 4 h, the tubes may be incubated overnight or longer to allow a positive reaction to develop.
3. Consult the table for guidance in interpretation of reactions.³

Production of Acid from:

Organisms	Base*	Dextrose	Lactose	Maltose	Sucrose
<i>N. gonorrhoeae</i>	–	+	–	–	–
<i>N. meningitidis</i>	–	+	–	+	–
<i>N. lactamica</i>	–	+	+	+	–
<i>N. sicca</i>	–	+	–	+	+

+ = Acid (yellow); – = no reaction (red)

*The base contains no carbohydrate. It is included to indicate a system failure such as a carry-over of carbohydrates from a primary isolation medium.

XI LIMITATIONS OF THE PROCEDURE

Additional tests should be performed for complete identification. Members of the genera *Kingella* and *Moraxella*, as well as commensal *Neisseria* species, may occasionally be isolated from patients with infections traditionally associated with *N. gonorrhoeae*.³

For identification, organisms must be in pure culture. Morphological, biochemical, and/or serological tests should be performed for final identification. Consult appropriate texts for detailed information and recommended procedures.⁵⁻⁷

XII PERFORMANCE CHARACTERISTICS

Rapid Fermentation Medium, Dextrose

Prior to release, all lots of Rapid Fermentation Medium, Dextrose are tested for performance characteristics. Representative samples of the lot are inoculated directly using fresh colony growth from the surface of a BBL Chocolate II Agar plate with *Neisseria gonorrhoeae* ATCC 43070, *N. meningitidis* ATCC 13090, *N. lactamica* ATCC 23970 and *N. sicca* ATCC 29193. Tubes are incubated at 35–37°C in an aerobic atmosphere and examined after 4 h. A positive reaction (color change from red to yellow) is observed with all organisms. Tubes may be incubated overnight to allow a stronger reaction.

Rapid Fermentation Medium, Lactose

Prior to release, all lots of Rapid Fermentation Medium, Lactose are tested for performance characteristics. Representative samples of the lot are inoculated directly using fresh colony growth from the surface of a BBL Chocolate II Agar plate with *Neisseria gonorrhoeae* ATCC 43070, *N. meningitidis* ATCC 13090, *N. lactamica* ATCC 23970 and *N. sicca* ATCC 29193. Tubes are

incubated at 35–37°C in an aerobic atmosphere and examined after 4 h. A positive reaction (color change from red to yellow) is observed with *N. lactamica* and a negative reaction (no color change) is observed with the remainder of the organisms. Tubes may be incubated overnight to allow a stronger reaction.

Rapid Fermentation Medium, Maltose

Prior to release, all lots of Rapid Fermentation Medium, Maltose are tested for performance characteristics. Representative samples of the lot are inoculated directly using fresh colony growth from the surface of a **BBL** Chocolate II Agar plate with *Neisseria gonorrhoeae* ATCC 43070, *N. meningitidis* ATCC 13090, *N. lactamica* ATCC 23970 and *N. sicca* ATCC 29193. Tubes are incubated at 35–37°C in an aerobic atmosphere and examined after 4 h. A positive reaction (color change from red to yellow) is observed with all organisms except *N. gonorrhoeae*, which is negative for maltose fermentation (no color change). Tubes may be incubated overnight to allow a stronger reaction.

Rapid Fermentation Medium, Sucrose


Prior to release, all lots of Rapid Fermentation Medium, Sucrose are tested for performance characteristics. Representative samples of the lot are inoculated directly using fresh colony growth from the surface of a **BBL** Chocolate II Agar plate with *Neisseria gonorrhoeae* ATCC 43070, *N. meningitidis* ATCC 13090, *N. lactamica* ATCC 23970 and *N. sicca* ATCC 29193. Tubes are incubated at 35–37°C in an aerobic atmosphere and examined after 4 h. A positive reaction (color change from red to yellow) is observed with *N. sicca* and negative reactions (no color change) are observed with the remainder of the organisms. Tubes may be incubated overnight to allow a stronger reaction.

XIII AVAILABILITY

Cat. No.	Description
221890	BBL™ Rapid Fermentation Medium, Base, Pkg. of 10 size K tubes
221891	BBL™ Rapid Fermentation Medium, Dextrose, Pkg. of 10 size K tubes
221893	BBL™ Rapid Fermentation Medium, Lactose, Pkg. of 10 size K tubes
221894	BBL™ Rapid Fermentation Medium, Maltose, Pkg. of 10 size K tubes
221895	BBL™ Rapid Fermentation Medium, Sucrose, Pkg. of 10 size K tubes

XIV REFERENCES

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