BBL™ CTA Medium™
BBL™ CTA Medium™ with Arabinose
BBL™ CTA Medium™ with Fructose
BBL™ CTA Medium™ with Mannitol
BBL™ CTA Medium™ with Sorbitol
BBL™ CTA Medium™ with Sucrose
BBL™ CTA Medium™ with Xylose

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QUALITY CONTROL PROCEDURES

I INTRODUCTION

CTA Medium™ (Cystine **Trypticase™** Agar Medium) is a simple, basic medium for the maintenance and detection of motility of a wide variety of microorganisms. With added carbohydrates, it is useful for the determination of fermentation reactions.

II PERFORMANCE TEST PROCEDURE

1. Inoculate representative samples with the cultures listed below.
   a. Loosen caps, boil the media in a boiling water bath* for approximately 2 min and tighten caps before cooling.
      ***NOTE:** Use of a microwave oven is not recommended.
   b. Inoculate the tubes using an inoculating needle to stab into the middle of the column of medium to a depth approximately one-half the total depth of the medium. For the *Neisseria* cultures, use a 0.01 mL calibrated loop and inoculate the surface of the medium using three or four colonies from a 24- to 48-h Chocolate II Agar plate. For the *Bacteroides* culture, use three or four colonies from a 24 – 48 h CDC Anaerobe Blood Agar plate. For the other test cultures, use 10^{-1} dilutions of 18- to 24-h **Trypticase** Soy Broth cultures.
   c. Incubate all tubes at 35 ± 2 °C. Incubate the *Neisseria* cultures with tightened caps. Incubate the remainder with loosened caps in an aerobic atmosphere with the exception of the *Bacteroides* culture which should be incubated in an anaerobic atmosphere (**GasPak™** EZ anaerobe system or equivalent).
2. Examine tubes after 18 – 24 and 42 – 48 h for growth and reactions.
3. Expected Results

Cultures of the following organisms are recommended for checking the performance of **CTA Medium** containing the various carbohydrates. The final colors produced should be compared with the color of the respective uninoculated medium.

Organisms	ATCC™	Base*	Arabinose	Fructose	Mannitol	Sorbitol	Sucrose	Xylose
Bacteroides fragilis	25285							A
Enterococcus faecalis	29212					A		
Escherichia coli	25922		A					
Listeria monocytogenes	19115	MK			MK	MK		
Morganella morganii	8019		K					
Neisseria gonorrhoeae	19424						K	K
Neisseria meningitidis	13090			K				
Neisseria sicca	29193			A				
Proteus vulgaris	8427		K					
Staphylococcus aureus	25923				A		A	

A = Acid (yellow); K = Alkaline or no reaction (red to reddish orange); M = Motile

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

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 7.6 ± 0.2.
4. Incubate uninoculated representative tubes at 20 – 25 °C and 30 – 35 °C and examine after 5 days for microbial contamination.

PRODUCT INFORMATION

IV INTENDED USE

CTA Medium is a culture medium for the maintenance of microorganisms. It is also used for the detection of bacterial motility and, with added carbohydrate, for fermentation reactions of fastidious microorganisms, i.e., *Neisseria*, pneumococci, streptococci and nonsporeforming anaerobes.

V SUMMARY AND EXPLANATION

CTA Medium was developed by Vera as a simple medium for the identification and maintenance of the gonococcus and other bacteria.[1]

CTA Medium without carbohydrates can be used for maintenance of cultures, including fastidious organisms, for extended periods when stored at appropriate temperatures.

CTA Medium with the appropriate carbohydrate is recommended for the differentiation of fastidious organisms by means of fermentation reactions. In the semisolid agar, acid reactions are easily detected because the acid formed is not immediately diffused throughout the entire culture. When no fermentable carbohydrate is present, most cultures show an alkaline shift.

Motility can be readily detected in the semisolid medium.[2] Stab cultures show growth out from the line of inoculation. Nonmotile organisms grow in the inoculated area, while the surrounding area remains clear.

BBL™ Taxo™ Carbohydrate Discs can conveniently be selected and added, as needed, to tubes of plain **CTA Medium** when fermentation reactions are to be determined.

For clostridia, bacilli, common micrococci, enteric bacilli, and other organisms not generally considered to be nutritionally fastidious, the use of **Trypticase** Agar Base is recommended instead of **CTA Medium**.

VI PRINCIPLES OF THE PROCEDURE

CTA Medium contains cystine and casein peptone to supply the nutrients necessary to support the growth of fastidious microorganisms.

Carbohydrate fermentation is detected by a visible color change of the medium due to the incorporation of the pH indicator dye, phenol red. When the carbohydrate present is metabolized by the organism, organic acids are produced and the medium becomes acidified. The peptone present in the medium, however, is also degraded by the bacteria present and yields substances which are alkaline in pH.

The phenol red indicator changes from reddish-orange to yellow when the amount of acid produced by carbohydrate fermentation is greater than the alkaline end products of peptone degradation.[3]

The color change with phenol red occurs around pH 6.8.

VII REAGENTS

CTA Medium

Approximate Formula* Per Liter Purified Water

L-Cystine	0.5 g
Pancreatic Digest of Casein	20.0 g
Agar	2.5 g
Sodium Chloride	5.0 g
Sodium Sulfite	0.5 g
Phenol Red	0.017 g

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

CTA Medium with Mannitol, Sorbitol, Sucrose or Xylose contains the above ingredients with, per liter, 5.0 g of the specified carbohydrate. **CTA Medium** with Arabinose or Fructose contains the above ingredients with, per liter, 10.0 g of the specified 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.

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

This product is not intended for use directly with specimens or mixed cultures. The organism to be tested must first be in pure culture.

IX PROCEDURE

Material Provided: CTA Medium or **CTA Medium** with Arabinose or **CTA Medium** with Fructose or **CTA Medium** with Mannitol or **CTA Medium** with Sorbitol or **CTA Medium** with Sucrose or **CTA Medium** with Xylose.

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

Test Procedure: Observe aseptic techniques.

1. Loosen caps, boil in a boiling water bath* for approximately 2 min, tighten caps and cool before use.
2. Remove fresh colony growth from the surface of a suitable culture medium, e.g., Chocolate Agar, not from a selective, primary isolation plate.[4]
3. For fermentation tests with members of the genus *Neisseria*, only the surface of the tubed medium is inoculated. For facultative organisms, such as streptococci and strictly anaerobic organisms, inoculate by stabbing the center of the medium with an inoculating needle to about half the depth of the medium.
4. Repeat for each tube to be inoculated.
5. Incubate at 35 ± 2 °C with loosened caps aerobically or anaerobically depending upon the organism being tested; *Neisseria* should be incubated with tight caps,[3,5] especially if tubes must be incubated in a CO_2 incubator,[6,7] or with loose caps in a non-CO_2 incubator.[8,9] Examine periodically up to 24 h for growth (turbidity), evidence of motility, and acid production (yellow color in upper layer of medium). A few strains may require incubation for up to 48 – 72 h.[10] The final colors produced should be compared with the color of the respective uninoculated medium.
6. Many fastidious organisms, including *Neisseria*, *Pasteurella*, streptococci, *Brucella*, corynebacteria and vibrios may be readily cultivated in **CTA Medium**, no added carbon dioxide, serum, or other enrichments being required.
7. For more rapid growth and also for more rapid fermentation reactions, anaerobic cultures preferably should be incubated in the presence of carbon dioxide as well as hydrogen or nitrogen. Some strict anaerobes fail to grow or grow poorly in the absence of carbon dioxide.

***NOTE:** Use of a microwave oven is not recommended.

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.

A single electrode of sufficiently small size to fit into the tubes should be used to determine the pH potentiometrically of tubed media. The tip of the electrode should be positioned in the central portion of the agar mass in semisolid media.

X RESULTS

A yellow color either in the upper one-third or throughout the medium indicates acid production; i.e., fermentation of the carbohydrate. A red (alkaline) to orange (neutral) color indicates that the carbohydrate has not been degraded and that only the peptone has been utilized. Inoculated plain **CTA Medium** (without carbohydrates) also exhibits a red to orange color.

Motile organisms show growth out from the line of stab-inoculation. Nonmotile organisms only grow along the stab line with the surrounding agar remaining clear.

XI LIMITATION OF THE PROCEDURE

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.[11-13]

XII AVAILABILITY

Cat. No.	Description
221632	**BD BBL™ CTA Medium™**, Ctn. of 100 size K tubes
297731	**BD BBL™ CTA Medium™** with Arabinose, Pkg. of 10 size K tubes
296001	**BD BBL™ CTA Medium™** with Fructose, Pkg. of 10 size K tubes
221639	**BD BBL™ CTA Medium™** with Mannitol, Pkg. of 10 size K tubes
221643	**BD BBL™ CTA Medium™** with Sorbitol, Pkg. of 10 size K tubes
221645	**BD BBL™ CTA Medium™** with Sucrose, Pkg. of 10 size K tubes
221647	**BD BBL™ CTA Medium™** with Xylose, Pkg. of 10 size K tubes

XIII REFERENCES

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