CYSTINE TRYPTIC AGAR (CTA)
w/ and w/o ASCITIC FLUID and CARBOHYDRATES

INTENDED USE
Remel Cystine Tryptic Agar w/ and w/o Ascitic Fluid and Carbohydrates are semisolid media recommended for use in qualitative procedures for the determination of fermentation reactions by fastidious organisms such as *Neisseria*, pneumococci, streptococci, and nonsporeforming anaerobes.

SUMMARY AND EXPLANATION
Cystine Tryptic Agar (CTA) was developed by Vera for the identification and maintenance of *Neisseria gonorrhoeae* and other bacteria.[1] CTA w/o Carbohydrates can be used for the maintenance of stock cultures while CTA w/ Carbohydrates differentiates fastidious organisms by means of fermentation reactions. Motility can be detected in CTA when the medium is inoculated by a stab line.[2] Herrmann recommended avoiding the addition of yeast extract, serum, or ascitic fluid to carbohydrate media.[3] However, to facilitate the fermentation reactions of *Neisseria* spp., the addition of ascitic fluid is recommended.[4]

PRINCIPLE
CTA medium contains cystine and casein peptone which supply essential nutrients for the growth of fastidious organisms. When the carbohydrate present is fermented, organic acids are produced and the medium becomes acidified. This results in the phenol red indicator changing from orange-red to yellow. The neutral orange color that results in CTA w/o Carbohydrate is caused by bacterial deamination of the peptone in the medium. Ascitic fluid may be added to CTA Agar as an enrichment to facilitate cultivation of fastidious organisms such as *Neisseria* spp.

REAGENTS (CLASSICAL FORMULAE)*
Base Medium:

Casein Peptone	20.0	g	Sodium Sulfite	0.5	g
Sodium Chloride	5.0	g	Phenol Red	17.0	mg
L-Cystine	0.5	g	Agar	2.5	g
			Demineralized Water	1000.0	ml

pH 7.3 ± 0.2 @ 25°C

These optional ingredients are available per liter of CTA medium:

Arabinose	10.0	g	Ribose	10.0	g
Dextrose	10.0	g	Salicin	10.0	g
Fructose	10.0	g	Sorbitol	10.0	g
Lactose	10.0	g	Sucrose	10.0	g
Maltose	10.0	g	Trehalose	10.0	g
Mannitol	10.0	g	Xylose	10.0	g

These optional ingredients are available per liter of CTA w/ Ascitic Fluid medium:

Dextrose	10.0	g	Maltose	10.0	g
Lactose	10.0	g	Sucrose	10.0	g

-*Adjusted as required to meet performance standards.

PROCEDURE
1. The performance of this medium is dependent on a properly prepared inoculum. Use pure cultures grown on nonselective medium to inoculate CTA medium. CTA Base w/o Carbohydrates should be inoculated and incubated in parallel with the fermentation test as a control to indicate a system failure, such as carryover of carbohydrate from a primary isolation medium.

    a. *Neisseria* spp.: Using a sterile 3 mm loop, remove a loopful of colonies from the surface of an 18-24 hour culture and inoculate the CTA medium a few millimeters below the surface. Alternatively, prepare a dense suspension of the test isolate in 0.5 ml of sterile saline (isotonic). With a Pasteur pipette, dispense 1 drop of the suspension onto the surface of the CTA medium, then stab the inoculum into the upper third of the medium.

    b. *Streptococcus* spp. (or similar organisms), anaerobes, or for motility testing: Inoculate by stabbing the center of the CTA medium with an inoculating needle to about half the depth of the medium.

2. Incubate in ambient air at 33-37°C for 24-72 hours with caps tightened. Do not incubate in CO_2. Examine tubes for a color change at periodic intervals for the first 24 hours of incubation. A few strains may require prolonged incubation, up to 72 hours.

INTERPRETATION OF THE TEST
Fermentation:

Positive Test -	Yellow color development
Negative Test -	Orange-red color

Motility:

Positive Test -	Growth out from the stab line of inoculation
Negative Test -	Growth along the stab line with the surrounding agar remaining clear

(Continued on back)

QUALITY CONTROL

All lot numbers of Cystine Tryptic Agar w/ and w/o Ascitic Fluid and Carbohydrates have been tested for performance and have been found to be acceptable. Testing of control organisms should be performed in accordance with established laboratory quality control procedures. Control organisms should be selected that demonstrate a positive and negative reaction for each carbohydrate tested. If aberrant quality control results are noted, patient results should not be reported.

LIMITATIONS

1. All isolates of *Neisseria gonorrhoeae* from medical legal cases should be confirmed with at least two methodologies.[5]
2. *Neisseria* spp. usually produces acid only in the upper third of the medium; a strong acid (yellow) throughout the medium may indicate contamination. Further testing following established laboratory procedures may be required for definitive identification.[6]

BIBLIOGRAPHY

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Refer to the front of Remel *Technical Manual of Microbiological Media* for **General Information** regarding precautions, product storage and deterioration, specimen collection, storage and transportation, materials required, quality control, and limitations.

IFU 60570, Revised November 10, 2010 Printed in U.S.A.

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