

Region V

Infertility Prevention Project

Self Study Manual

Developed by the Region V

Infertility Prevention

Training Committee

June 2006



HEALTH CARE EDUCATION & TRAINING, INC.

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SELF STUDY MANUAL

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TABLE OF CONTENTS

- **INTRODUCTION AND INSTRUCTIONS Module 1**
- **CHLAMYDIA MEDICAL OVERVIEW Module 2**
- **GUIDELINES FOR SCREENING & SPECIMEN COLLECTION... Module 3**
- **LABORATORY Module 4**
- **CASE STUDIES Module 5**
- **DIAGNOSIS AND TREATMENT OF CHLAMYDIA Module 6**
- **DATA Module 7**
- **CLIENT EDUCATION AND COUNSELING Module 8**
- **QUALITY ASSURANCE Module 9**
- **GLOSSARY Module 10**
- **APPENDIX**
 - Knowledge Assessment Test & Answer Sheet
 - Employee Checklist
 - Manual Evaluation
 - Nursing Continuing Education Application Form
 - Educational Resources
 - Training Committee Members
 - References

COURSE OBJECTIVES

Upon completion of this course the reader should be able to:

- Identify the components of a quality assurance programs for *Chlamydia trachomatis* (CT) screening in the clinic setting.
- Discuss the epidemiology and pathogenesis of CT.
- Identify the clinical presentation and diagnosis of CT genital infections in men and women.
- Describe the recommended screening criteria.
- Describe correct specimen collection techniques and common problems of specimen collection and handling.
- Discuss the basic principles, utility and interpretation of commonly used laboratory tests for the diagnosis of CT infection and common reasons for specimen rejection.
- Describe treatment recommendations and appropriate regimens for CT based on patient profile.
- List elements and processes of data collection and reporting activities.
- Identify essential information that must be presented to the client regarding partner referral and behavior change strategies.

HISTORY & ORGANIZATION OF THE REGION V INFERTILITY PREVENTION PROJECT

WHAT IS THE INFERTILITY PREVENTION PROJECT?

In 1993 Congress appropriated funds to the Centers for Disease Control and Prevention (CDC) to begin a national STD-related Infertility Prevention Program. The program was designed to improve screening, surveillance and treatment of the infection, *Chlamydia trachomatis* (CT), in the United States. By 1996 the CDC had contracted with all states for demonstration level health funding to provide tests/treatment for chlamydia in selected family planning and sexually transmitted disease clinics.

Approximately 75% of infected women and 50% of infected men have no symptoms of CT, and therefore, may not seek health care until severe health problems occur. When diagnosed CT can be easily treated and cured. Untreated CT can cause severe and costly reproductive and health problems including pelvic inflammatory disease (PID), which frequently results in chronic pelvic pain and is linked to infertility and ectopic pregnancy. CT is one of the major causes of tubal infertility in the United States.¹

WHO IS INVOLVED IN THE REGION V INFERTILITY PREVENTION PROJECT?

The six states in Region V, Illinois, Indiana, Michigan, Minnesota, Ohio, and Wisconsin, have been working collaboratively since 1995 through the RVIPP Advisory Committee. Thirty members from the six states worked jointly to develop screening criteria, collect data, establish volume purchasing, exchange information and research, and set regional objectives. The members represent the fields of family planning, STD, maternal and child health, public health laboratories, and epidemiology. Each state has an infertility prevention state alliance that relays recommendations to the RVIPP Advisory Committee and in turn, receives guidance from the RVIPP. Regional activities are coordinated through Health Care Education and Training, Inc. (HCET), a non-profit organization that provides training, technical assistance, and infrastructure development on issues of women's health within the region.

WHAT ARE THE HEALTH CARE COSTS OF UNTREATED CHLAMYDIA?

According to the Alan Guttmacher Institute in 2002, the population of Region V consisted of 11,581,600 women of childbearing age with 6,091,910 of these women in need of contraceptive services and supplies. Childbearing age is defined here as 13-44 years of age and need is defined as a person living at or below 100% of the poverty level.²

There were 177,069 cases of CT reported in 2004 in Region V and 929,462 reported nationally.³ Up to 40% of women with untreated CT infection will develop PID.¹ CDC estimates the health care costs of untreated chlamydia to be more than \$2 billion annually in the United States, while screening and treatment programs cost \$175 million annually. That means that every dollar spent toward screening and treatment will save \$12 in complications that result from untreated CT.⁴

CHLAMYDIA TRACHOMATIS

INTRODUCTION

Chlamydia trachomatis (CT) is the most common bacterial infection of the genital tract in most developed countries and in many developing countries. An estimated 2.8 million new infections occur annually in the United States.⁵ Acute symptomatic infections result in substantial costs, both in terms of morbidity and dollars. However, untreated asymptomatic infections may cost even more in terms of sequelae including infertility and ectopic pregnancy. In the developing world, certain strains also cause trachoma, one of the most common, potentially preventable, causes of blindness.

ETIOLOGY

CT is an obligate intracellular bacterium with a two phase life cycle. The infectious form, elementary bodies (EBs), attaches to and enters the host cell. After entering the cell, they reorganize into the metabolically active and replicative reticulate bodies (RB). Using host-derived adenosine triphosphate (ATP) as an energy source, the RBs divide by binary fission producing up to several hundred progeny in a large cytoplasmic inclusion within the cell. After replication the RBs reorganize into the infectious EBs, which are released by the host cell. The reproductive cycle takes 48 to 72 hours in tissue culture; however, the reproductive cycle probably takes longer in the body.

EPIDEMIOLOGY

Where CT has been reported and studied in comparison with gonorrhea (GC), the incidence of CT exceeds that of GC. Conservative estimates indicate that 1 in every 20 sexually active women of childbearing age and 1 in every 10 adolescent girls are infected with CT.⁶ The prevalence of CT infection in men ranges from 4% to 10% in asymptomatic populations and from 15% to 20% in young men attending STD clinics.^{7,8} Incidence rates for CT are highest in the 15 -19 year age range and 74% of reported positives are in individuals under 25 years.⁹ The chlamydial infection rate increases through the teenage years and into the early twenties and then declines.⁸ To some extent these trends parallel rates of high risk sexual activity (i.e. unprotected sex, multiple partners, etc.), but even if this is taken into account, it still appears that the rate of infection falls with age suggesting the possibility of acquired immunity.

Populations at an increased risk of infection include men and women who are sexually active under the age of 25, use condoms inconsistently, and/or have multiple partners.¹ Heterosexual women and men are at higher risk than their homosexual counterparts. The risk of acquiring chlamydial infection from an infected partner has been difficult to define because infection is so often asymptomatic and data from single encounters with an infected person are sparse. However, the risk of infection has been found as high as 65% for a woman who has had multiple contacts with an infected partner.¹⁰ It is important to note that people with GC infections are frequently co-infected with CT; in some populations the reverse is true as well.

Clinical presentations which identify women infected with or highly likely to be diagnosed with CT include:

- pelvic inflammatory disease (PID)
- gonorrhea (GC)
- urethral syndrome (pain on urination with inflammation but no significant number of bacteria culturable from the urine)

Men are likely infected with CT if they present with either of the following:

- urethritis (gonococcal or nongonococcal)
- epididymitis

PATHOGENESIS

In women the initial site of infection is usually the endocervical columnar epithelial cells. Due to the presence of columnar epithelia on the ectocervix (ectopia) in adolescents and in oral contraceptive users, these women are more susceptible to infection with CT. Squamous epithelia, such are present in the vagina, are resistant, and thus adult women are less likely to develop chlamydial vaginitis. However, the transitional cell epithelia of the prepubertal female is susceptible and chlamydial vaginitis and urethritis may occur.¹

Infection leads to cervicitis in most if not all women. Cervicitis may resolve spontaneously or leave a low-grade chronic infection with minimal signs of inflammation. However, infection frequently ascends

to the upper genital tract to involve the endometrium and fallopian tubes producing endometritis and chronic salpingitis. The severity and the chronicity of chlamydial infections appear to be highly variable.

Infection in women is typically subclinical or asymptomatic. However, if left untreated, infection may be associated with the same sequelae as clinically diagnosed salpingitis, that is, inflammation and scarring of the fallopian tubes leading to pelvic pain, fever, tubal obstruction, infertility, and high risk of having ectopic pregnancy. Acute chlamydial salpingitis is characterized by mucosal edema and inflammatory changes that may involve all layers of the tubes. When symptomatic, acute endocervical infection with CT is frequently characterized by purulent discharge, which at times may be perceived as a vaginal discharge.

In men infections usually remain localized to the urethra but may spread retrograde to cause epididymitis or perhaps prostatitis. A new infection usually results in an acute inflammatory response similar to that associated with gonococcal urethritis but often with fewer white cells and less discharge. The natural course of untreated infection in men is unknown although some appear to develop a chronic low-grade urethritis. As many as 50% of sexually active men have asymptomatic or minimally symptomatic chlamydial infections, these infections may resolve spontaneously, but the duration of subclinical infection is unknown.¹

COMPLICATIONS OF INFECTION IN FEMALES

Urethral Syndrome

Chlamydial infection occasionally causes urethral syndrome, which is defined as dysuria with or without pyuria in the absence of significant bacteriuria. CT can also be isolated from the urethra of women without urethral symptoms and negative endocervical cultures. It is believed that these women have endocervical infection but that it is not detected by culture.

Endometritis

Endometritis is common in women with chlamydial infection. Endometritis may be symptomatic or asymptomatic. It persists despite shedding of the endometrium with menses. Symptoms usually consist

of low-grade fever, abdominal pain, cramping and bleeding between menstrual periods. A histopathologic or microbiological diagnosis may be made by endometrial biopsy. However, the organism recovered may represent contamination from the endocervix. Testing of the endometrium may be more sensitive than testing of the endocervix for diagnosis of chlamydial infection; however, endometrial sampling is not a routine procedure.

Salpingitis and Acute Pelvic Inflammatory Disease

CT may spread from the endometrium to the fallopian tubes to produce acute salpingitis or PID. Symptoms include abdominal or pelvic pain, fever, nausea, vomiting or other systemic manifestations. Physical examination reveals tenderness on movement of the cervix (the chandelier sign) and in the adnexa or uterine fundus. Occasionally, right upper quadrant pain dominates the clinical picture as a result of perihepatitis (Fitz-Hugh-Curtis syndrome) in which adhesions to the liver capsule are seen at laparoscopy. Acute symptomatic chlamydial PID is more frequent in younger women. More commonly, chlamydial PID is clinically chronic and associated with mild to moderate abdominal pain and less impressive tenderness on pelvic examination. However, such women may have significant tubal inflammation and adhesions at laparoscopy. The nature of the symptoms and the paucity of physical findings may delay diagnosis.

The chance of becoming infertile after symptomatic PID increases with the severity and number of episodes and ranges from 5.8% to 60%.^{11,12} However, most women who have tubal infertility have no history of salpingitis. They may have serologic evidence of a prior chlamydial infection, presumably reflecting subacute or chronic salpingitis. In some cases they appear to have persistent infection since CT is recovered from the fallopian tubes of about 15% of infertile women undergoing microtuboplasty for tubal infertility.¹³ The likelihood of infertility after asymptomatic untreated infection is unknown.

INFECTION IN NEONATES

An infant born to a woman with chlamydial infection has about a 55% chance of being infected, but many are asymptomatic. About 20-50% of infants born to infected women develop neonatal inclusion conjunctivitis and 10-20% of infants develop pneumonia.¹⁴⁻¹⁶ Conjunctivitis typically presents 5 to 12 days after birth with erythematous conjunctivae and an ocular discharge that may be purulent. In infants who have received topical antibiotic prophylaxis at birth this tends to develop later. Spontaneous resolution can occur without significant sequelae; however, conjunctival scarring has been reported, and blindness can occur if not aggressively treated.

Pneumonia usually presents 3-11 weeks after delivery with a cough and tachypnea with little or no fever. CT can be recovered from sputum or the nasopharynx. If untreated the course is usually protracted with gradual resolution of symptoms over several weeks; however, it can be life-threatening. Although chlamydial infections in infants may be relatively benign, there is some evidence that pneumonia in the first 6 months of life may be associated with reactive airway disease in childhood.

INFECTION IN MALES

Most men with symptomatic CT infection present with pain on urination and discharge that ranges from clear to grossly purulent. The discharge is frequently noticed when it stains underwear and typically begins seven to ten days after a new sexual contact. Physical examination usually reveals a discharge, and “milking” or “stripping” the urethra (i.e. proximal to distal massage of the ventral penis) may increase detection. Inguinal lymph nodes are typically not enlarged or tender.

A Gram stain of the exudate shows nongonococcal urethritis (NGU), i.e. the presence of neutrophils and no intracellular diplococci (in the absence of concomitant gonococcal infection). In most studies CT has been associated with 30-50% of all cases of NGU.¹⁷⁻¹⁹ No clinical features reliably distinguish chlamydial and nonchlamydial NGU. Symptoms and incubation periods are similar (7-21 days). However, CT co-infection is found in approximately 20-40% of men infected with GC.²⁰⁻²²

Other sexually transmitted pathogens that may cause non-chlamydial NGU include *Mycoplasma hominis*, *Ureaplasma urealyticum*, Herpes simplex viruses (HSV), and *Trichomonas vaginalis*. Urethritis may also occur in association with urinary tract infections, prostatitis, urethral strictures, phimosis, Reiter's syndrome, or after instrumentation.

Epididymitis

Epididymitis presents as unilateral pain and swelling of the epididymis, inguinal pain and occasionally scrotal erythema or edema. Expressible urethral discharge may be present and is particularly common in men under the age of 35. Older men with epididymitis are more likely to be infected with coliform bacteria like *Escherichia coli* (E. coli) or *Pseudomonas aeruginosa*. GC also causes epididymitis in sexually active men.

Other Manifestations in Males

Prostatitis has been reported in association with chlamydial infection but it is unclear whether it is a manifestation of infection. Reiter's syndrome is an immune-mediated systematic illness that occurs more often in men than women. It is characterized by arthritis, conjunctivitis, and urethritis which occur about one month after CT or other genital infections.

RARE MANIFESTATIONS

C. trachomatis has been associated with endocarditis, pleuritis, pneumonitis, mediastinal and supraclavicular lymphadenopathy, hepatitis, fever of unknown origin and dilated cardiomyopathy.

SUMMARY: CHLAMYDIA MEDICAL OVERVIEW

PATHOPHYSIOLOGY

- CT is an obligate, intracellular bacterium that replicates in 48-72 hours and incubates in 1-3 weeks.
- It preferentially infects columnar or transitional epithelia, such as the urethra or endo cervix.
- Infections are often chronic, asymptomatic and reinfection is common.

CLINICAL PRESENTATION

Women

- 1/2 – 2/3 asymptomatic
- Cervicitis
- PID
- Perihepatitis (Fitz-Hugh-Curtis Syndrome)
- Urethritis – Usually asymptomatic
- Vertical Transmission (pneumonia, conjunctivitis) to newborn

Men

- Urethritis
- Reiter's Syndrome
- Proctitis
- Epididymitis

MEDICAL OVERVIEW QUIZ

- 1. All of the following statements concerning *Chlamydia trachomatis* (CT) are true EXCEPT:**
 - A. It is the most common bacterial infection of the genital tract in most developed countries.
 - B. Ten million cases of CT genital infection occur annually in the United States.
 - C. Sequelae of asymptomatic infections include infertility and ectopic pregnancy.
- 2. Which of the following women would be considered most at risk for CT?**
 - A. A 17-year-old woman who has been with her new partner for the past two months.
 - B. A 23-year-old woman in a mutually monogamous relationship who has an IUD.
 - C. A 25-year-old single woman who uses a diaphragm for contraception.
- 3. In women, the initial site of chlamydial infection is usually?**
 - A. Cervical columnar epithelia
 - B. Cervical squamous epithelia
 - C. Uterine endometrial tissue
 - D. Vaginal mucosal epithelia
- 4. Most women who have tubal infertility have had previously diagnosed salpingitis.**
 - A. True
 - B. False
- 5. A woman who has had three episodes of PID is no more likely to become infertile than a woman who has had one episode of PID.**
 - A. True
 - B. False

Answers:

1. B
2. A
3. A
4. B
5. B

REGION V SCREENING CRITERIA RECOMMENDATIONS

Development of Screening Criteria

During 1997 universal testing of *Chlamydia trachomatis* (CT) was implemented in study, sentinel, and clinic sites in Illinois, Indiana, Michigan, Minnesota, Ohio and Wisconsin. All persons who tested positive were treated if located. The clinic sites were strategically located to encompass rural/urban, multicultural populations and various income brackets. Family planning and sexually transmitted disease clinics participated in the study. Behavioral, clinical, laboratory and demographic data were collected for one year using laboratory requisition forms and patient completed questionnaires. In 1998 the Region V Epidemiologist, John Pfister, analyzed the data collected by each state and presented the findings at a regional meeting of program collaborators. The data were analyzed by multiple variables looking for significant trends in prevalence. A computer application was developed so that each state could input their budget and prevalence data to determine the most appropriate screening criteria.

Region V screening criteria recommendations are formulated by evidence-based data gathered in this region. Each state has analyzed state specific data and developed recommended screening criteria based on the state's data and budget availability to purchase tests.

In the 2002 Sexually Transmitted Diseases Treatment Guidelines, the Centers for Disease Control & Prevention (CDC) expanded their screening recommendations to include annual screening of sexually active adolescents (19 years and under), young adult women (20 – 24 years), and other women with a risk factor for CT. This recommendation is not constricted by state budget limitations. In addition, the CDC recommends re-screening 3-4 months after treatment for a CT infection. This is not as a “test of cure” but due to the high risk of re-infection often caused by the patient's sex partner not being treated and/or the patient continuing high risk behavior following treatment. Some experts especially recommend re-screening adolescents due to the high rate of re-infection in this population.

The Region V Screening Criteria Recommendations

Family Planning Clinic

Females: Women should be screened who meet at least one of the following criteria:

- Age (based upon data indicators and resources) < 19; < 19 biannually; or < 23
- Signs or symptoms
- STD Contact
- Sex Partner Risk (new sex partners, multiple sex partners, sex partner with other partner)
- Prior STD History

Males: Dependent upon agency resources

STD Clinic

Females: Women should be screened who meet at least one of the following criteria:

- Age (based upon data indicators and resources) < 21; or < 27
- Signs and symptoms

- STD Contact
- Sex Partner Risk (new or multiple partners, sex partner with other partners)

Males: All

SPECIMEN COLLECTION INSTRUCTIONS BY TEST TYPE

DIRECT FLUORESCENT ANTIBODY (DFA)

Female Cytobrush (preferred female collection device for DFA)

1. Remove excess mucus using cotton or dacron swab. Discard swab.
2. Insert cytobrush into endocervical canal past squamocolumnar junction. Leave in place 2-5 seconds.
3. Rotate brush one full turn (360°). Withdraw brush without touching vaginal surfaces.
4. Place portion of cytobrush containing the specimen across the center of the slide well.
5. Rotate and twist the brush, moving the brush back and forth across the slide well. Liquid clinging to the brush will disperse cells across the slide well.
6. Check coverage.
7. Label slide.
8. Air dry completely.
9. Fix with methanol.

Female Swab

1. Remove excess mucus using cotton or dacron swab.
2. Insert second swab into endocervical canal until the tip is not visible.
3. Rotate swab 360° for 5-10 seconds inside endocervical canal. Withdraw swab without touching vaginal surfaces.
4. Prepare slide immediately by rolling one side of swab over top half of slide well and other side of swab over bottom half of slide well.
5. Label slide.
6. Air dry completely.
7. Fix with methanol.

Male Swab

1. Collect all other samples first. Preferably no urination one hour prior to collection.
2. Using manufacturer's male collection kit swab, insert into urethra a minimum of 2-4 cm.
3. Rotate at least one complete revolution for 2-5 seconds and withdraw swab.
4. Prepare slide immediately by rolling one side of swab over top half of slide well and other side of swab over bottom half of slide well.
5. Label slide.
6. Air dry completely.
7. Fix with methanol.

SYVA MICRO TRAK - ENZYME IMMUNOASSAY (EIA)

Female Swab

1. Cleanse ectocervix lightly with an extra swab. (You may use one not included in the kit).
2. Insert kit dacron swab into endocervical canal until tip is not visible.
3. Rotate swab in the canal for 15-30 seconds.
4. Remove swab, being careful not to touch vaginal surfaces.
5. Place swab immediately in labeled transport tube with bar code attached.
6. Break off shaft of swab at score line.
7. Insert the end of the swab into the center hole of the cap.
8. Cap tube tightly.

Male Swab

1. Client should not urinate for one hour before testing.
2. Express and discard any pus or exudate before testing.
3. Insert small swab with wire handle 2- 4 cm into the urethra.
4. Gently rotate swab for 15 seconds.
5. Remove swab.
6. Place swab immediately in labeled transport tube with bar code attached.
7. Break swab handle.
8. Insert the end of the swab into the center hole of the cap.
9. Cap tube tightly.

Remember: 1) columnar epithelial cells must be present for reliable results therefore, do not sample only discharge/exudate, 2) use ONLY the swabs in the kit to collect the specimens, 3) break the kit swab at the score line otherwise the cap may not secure tightly, causing the specimen to leak/become inadequate for testing, and 4) if transporting by U.S. Post Office, please confirm shipping requirements for biological specimens, except DFA.

GEN-PROBE - NUCLEIC ACID PROBE

Female Swab (Collect specimen after a PAP smear, if being done, and/or wet mount)

1. Remove excess mucus from cervical and surrounding mucosa using one of the swabs provided. Discard the cleaning swab.
2. Insert second swab from collection kit 1 - 1 1/2 cm into endocervical canal.
3. Rotate swab 10-30 seconds in endocervical canal to ensure adequate sampling.
4. Withdraw swab carefully; avoid any contact with vaginal mucosa.
5. Immediately place swab in specimen collection kit transport tube.
6. Break swab shaft at scoreline.
7. Replace cap tightly and label tube.
8. Transport to the laboratory at 2° C to 25° C as soon as possible after collection. Specimens must reach the laboratory within 7 days after collection or they will be unsatisfactory for testing.
9. If transporting by U.S. Post Office, please confirm shipping requirements for biological specimens.

Male Swab

1. Patient should not have urinated for at least 1 hour prior to sample collection.
2. Insert swab from the collection kit 2-4 cm into urethra.
3. Rotate swab gently at least one full rotation for 2-3 seconds. Use sufficient pressure to ensure swab comes into contact with all urethral surfaces.
4. Immediately place swab in specimen collection kit transport tube.
5. Break swab shaft at scoreline.
6. Replace cap tightly and label tube.
7. Transport to the laboratory at 2°C to 25° C as soon as possible after collection. Specimens must reach the laboratory within 7 days after collection or they will be unsatisfactory for testing.
8. If transporting by U.S. Post Office, please confirm shipping requirements for biological specimens.

Remember: 1) columnar epithelial cells must be present for reliable results, 2) use ONLY the swabs in the kit to collect the specimens, 3) do not collect specimens from females with swabs and transports intended for males, 4) break the kit swab at the score line otherwise the cap may not secure tightly, causing the specimen to leak/become inadequate for testing.

POLYMERASE CHAIN REACTION (PCR)

Female Swab

1. Remove excess mucus using one of the large swabs included in specimen collection kit. Discard swab.
2. Insert other large swab into endocervical canal until the tip is not visible.
3. Rotate the swab 360° for 5-10 seconds inside the endocervical canal. Withdraw swab without touching vaginal surfaces.
4. Place swab in Specimen Transport Media tube, vigorously swirl or agitate the swab in the liquid for 15 seconds.
5. Replace cap tightly and label tube.

Male Swab

1. Preferably no urination one hour prior to collection.
2. Using manufacturer's male collection kit swab, insert into urethra a minimum of 2-4 cm.
3. Rotate at least one complete revolution for 2-5 seconds and withdraw swab.
4. Place swab in the Specimen transport Medium tube, vigorously swirl or agitate the swab in the liquid for 15 seconds.
5. Replace cap tightly and label tube.

Male Urine

1. Collect all other samples first. Preferably no urination one hour prior to collection.
2. Collect first 15-20 ml urine in collection cup.
3. Replace cap tightly and label cup.

Remember: (1) columnar epithelial cells must be present for reliable results, (2) use ONLY the swabs in the kit to collect the specimens, (3) do not collect specimens from females with swabs and transports intended for males, (4) break the kit swab at the score line otherwise the cap may not secure tightly, causing the specimen to leak/become inadequate for testing.

APTIMA COMBO 2 ASSAY – TRANSCRIPTION MEDIATED AMPLIFICATION (TMA)

Female Swab

1. Remove excess mucus from cervical os and surrounding mucosa using cleaning swab in package (white shaft swab in package with red printing). Discard this swab.
2. Insert specimen collection swab (blue shaft swab in package with green printing) into endocervical canal.
3. Gently rotate swab clockwise for 10 to 30 seconds in the endocervical canal to ensure adequate sampling.
4. Withdraw swab without touching vaginal surfaces.
5. Remove cap from swab specimen transport tube and immediately place specimen collection swab into specimen transport tube.
6. Carefully break swab shaft at scoreline; use care to avoid splashing contents.
7. Re-cap swab specimen transport tube tightly.

See Specimen Transport and Storage below.

Male Swab

1. Patient should not have urinated one hour prior to specimen collection.
2. Using manufacturer's male collection kit swab (blue shaft swab in package with green printing), insert into urethra 2-4 cm.
3. Rotate swab clockwise for 2-3 seconds in urethra to ensure adequate sampling.
4. Withdraw swab carefully.
5. Remove cap from swab specimen transport tube and immediately place specimen collection swab into specimen transport tube.
6. Carefully break swab shaft at scoreline; use care to avoid splashing contents.
7. Re-cap swab specimen transport tube tightly.

See Specimen Transport and Storage below.

Specimen Transport and Storage

After collection, transport and store swab in swab specimen transport tube at 2°C to 30°C until tested. Specimens must be assayed with the GEN-PROBE APTIMA Combo 2 Assay within 60 days of collection. If longer storage is needed, freeze at -20°C to -70°C for up to 90 days after collection.

Male and Female Urine

1. Patient should not have urinated for at least one hour prior to specimen collection.
2. Direct patient to provide first-catch urine (approximately 20 to 30 mL of initial urine stream) into urine collection cup free of any preservatives. Collection of larger volumes of urine may result in specimen dilution that may reduce test sensitivity. Female patients should not cleanse labial area prior to providing specimen.

3. Remove cap from urine specimen transport tube and transfer 2 ml. Of urine into urine specimen transport tube using disposable pipette provided. The correct volume of urine has been added when fluid level is between black fill lines on urine specimen transport tube label.
4. Re-Cap urine specimen transport tube tightly. This is known as the “processed urine specimen”.
5. See Specimen Transport and Storage below.

Specimen Transport and Storage

After collection, transport the processed urine specimens in the GEN-PROBE APTIMA COMBO 2 Assay urine specimen transport tube at 2°C to 30°C and store at 2°C to 30°C until tested. Processed urine specimens should be assayed with the APTIMA Combo 2 Assay within 30 days of collection. If longer storage is needed, freeze at -20°C to -70°C for up to 90 days after collection.

Urine samples that are still in primary collection containers must be transported to lab at 2°C to 30°C. Transfer urine sample into APTIMA Combo 2 Assay urine specimen transport tube within 24 hours of collection. Store at 2°C to 20°C and test within 30 days of collection.

BDProbeTec- BD Biosciences

Female Swab-Endocervical Swab Specimen Collection

1. Remove excess mucus from the cervical os with the large-tipped cleaning swab provided in the kit and discard
2. Insert 2nd swab from kit into the cervical canal and rotate for 15 – 30 seconds.
3. Remove the swab carefully. Avoid contact with the vaginal mucosa.
4. Immediately place the cap/swab into the transport tube. Make sure the cap is tightly secured to the tube.
5. Label the tube with patient information and date/time collected.

Male Swab Urethral Swab Specimen Collection

1. Insert the Mini-Tip swab from the kit 2-4 cm into the urethra and rotate for 3-5 seconds.
2. Withdraw the swab and place the cap/swab into the transport tube. Make sure the cap is tightly secured to the tube.
3. Label the tube with patient information and date/time collected.

Swab Storage and Transport:

The Culturette Direct Collection swab and the Mini-Tip Culturette Direct swab must be stored and transported to the laboratory and/or test site at 2-27°C within 4-6 days of collection. Storage up to 4 days has been validated with clinical specimens. Storage up to 6 days has been demonstrated with seeded specimens. Note: If specimens cannot be transported directly to the testing laboratory under ambient temperatures (15-27°C) and must be shipped, an insulated container with ice should be used with either an overnight or 2-day delivery vendor.

Remember: (1) columnar epithelial cells must be present for reliable results, (2) use ONLY the swabs in the kit to collect the specimens

BDProbeTec- BD Biosciences

Urine Specimen Collection

Urine Processing Pouches (UPPs)

The following Collection/Storage/Transport instructions apply when no UPP is added at the collection site.

1. The patient should not have urinated for at least one hour prior to specimen collection.
2. Collect specimen in a sterile, plastic, preservative-free specimen collection cup.
3. The patient should collect the first 15 – 20 ml. of voided urine (the first part of the stream – NOT MIDSTREAM).
4. Label with patient identification and date/time collected.

Urine Storage and Transport

1. Store and transport urine specimens to the test site at 2 - 8°C within 4-6 days of collection.
2. Add the UPP to the urine specimen collection cup. Wear gloves when handling the UPP and urine specimen.
3. Cap the collection cup and swirl gently to ensure the UPP is completely submerged in urine.
4. The UPP must be in contact with the urine specimen for at least 2 hours prior to the processing.
5. Do not freeze the specimen.

NOTE: Specimens must be shipped in an insulated container with ice, using either an overnight or 2-day delivery vendor. Storage up to 4 days has been validated with clinical specimens. Storage up to 6 days has been demonstrated with seeded specimens.

Urine Specimen Collection

The following Collection/Storage Transport instructions apply when UPP is added at the collection site.

1. The patient should not have urinated for at least one hour prior to specimen collection.
2. Collect specimen in a sterile, plastic, preservative-free specimen collection cup.
3. The patient should collect the first 15 – 20 ml. Of voided urine (the first part of the stream – NOT MIDSTREAM).
4. Immediately add the UPP to the specimen collection cup. Wear gloves when handling the UPP and urine specimen.
5. Cap collection cup and swirl gently to ensure that the UPP is completely submerged in urine.
6. Label with patient identification and date/time collected.

NOTE: Specimens cannot be transported directly to the testing laboratory under ambient temperatures (15-27°C) and must be shipped, an insulated container with ice should be used with either an overnight or 2-day delivery vendor. Storage up to 4 days has been validated with clinical specimens. Storage up to 6 days has been demonstrated with seeded specimens.

1. Store and transport urine specimens containing a UPP to the laboratory or test site at 2 - 8°C within 4-6 days of collection or at 15-27°C within 2 days of collection.
2. Do not freeze the urine specimen.
3. The UPP must be in contact with the urine specimen for at least two hours prior to processing.

CRITERIA FOR SCREENING AND SPECIMEN COLLECTION OVERVIEW QUIZ

- 1. Which of the following asymptomatic 26-year-old females meets the criteria for *Chlamydia trachomatis* screening?**
 - A. Monogamous relationship for six months, pregnant, having an induced abortion.
 - B. Monogamous relationship for one year, does not use a barrier contraceptive method.
 - C. New partner in last three months, uses barrier contraception on a regular basis.
- 2. A 17-year-old woman who *was* treated for *Chlamydia trachomatis* four months ago has returned for her annual exam. The reason that another CT test is recommended at this visit is:**
 - A. Teenagers often do not complete their medication.
 - B. She may have a resistant strain of chlamydia.
 - C. There is a high rate of re-infection among teenage women.
- 3. Which of the following statements concerning the collection of cervical specimens for *Chlamydia trachomatis* testing is true?**
 - A. Excess mucous should be removed from the cervix prior to obtaining the specimen.
 - B. The swab should be rotated in the endocervix for *five* seconds to assure adequate sampling.
 - C. The specimen for *Chlamydia trachomatis* testing should be obtained before any other cervical specimen.
- 4. Which of the following statements concerning the collection of male urethral specimens for *Chlamydia trachomatis* testing is true?**
 - A. Insert the swab 2-4 cm into the urethra to obtain the specimen.
 - B. Rotate the swab for 30 seconds.
 - C. Ask the patient to urinate prior to obtaining the specimen.
- 5. Which of the following statements concerning the swabs used to obtain the specimens for *Chlamydia trachomatis* testing is true?**
 - A. A swab with any blood on it should be discarded and another cotton swab should be used.
 - B. Excess mucous should be wiped off the swab before placing in the transport tube.
 - C. Only the swabs that are supplied with the test kit can be used to collect specimens.

Answers:

1. A
2. C
3. A
4. A
5. C

INTRODUCTION TO LABORATORY METHODS

Numerous laboratory tests exist for diagnosing *Chlamydia trachomatis* (CT) infections. Understanding a little about the various tests and how they work will enable you to submit the best specimens and properly interpret results. Based on performance and cost issues, laboratory methods for CT can be divided into the following categories listed below. A chart listing highlights and specific points of interest for each type of test can be found on pages 4-3 and 4-4.

- A. *Culture*: Isolation and propagation of chlamydial organisms using cell cultures was the first practical method for laboratory diagnosis and is still in use today though usually recommended only under special circumstances. No non-culture method can match the specificity of culture, which is considered to be virtually 100% specific; thus, almost every positive is a true positive. It is this unparalleled specificity and corresponding high positive predictive value (PPV) that makes culture desirable in cases such as suspected sexual abuse, where a false-positive result can be especially devastating. Unfortunately, culture can lack sensitivity which means it is often falsely negative. This lack of sensitivity combined with the expense, rigorous specimen handling requirements, long time to results and scarce availability of culture limits its usefulness in routine management of individuals at-risk.
- B. *Non-Culture, Non-Amplified Methods*: This is the largest and most diverse group of CT tests and contains several subgroups, the largest of which is the Antigen Detection subgroup. This subgroup includes Enzyme Immunoassays of several different formats (micro-plate, automated, and rapid point-of-care tests) and Direct Fluorescent Antibody (DFA.) Nucleic acid probe (NAP) comprises the second subgroup. This subgroup is made up of only one test, the Gen Probe PACE and PACE2 assays, probably the most commonly used non-amplified CT test. Although NAP utilizes nucleic acid hybridization, it is **not** an amplified test, and performance and cost issues are similar to that of Antigen Detection methods. The third subgroup is also comprised of one test, the Digene Hybrid Capture assay, which incorporates “signal amplification” to enhance sensitivity over EIA and NAP type methods though it does not achieve the sensitivity of “target amplification” tests described below. This relatively new test is being touted as a less-expensive alternative to Nucleic Acid Amplification Test (NAAT), but it is not really considered a NAAT.
- C. *Nucleic Acid Amplification Tests*: The newest and most rapidly expanding category of CT tests and by far the most sensitive, NAAT’s are rapidly becoming the chlamydia tests of choice in many settings. The improved performance does come at a price as NAAT’s are still significantly more expensive than other test methods. However, with the number of commercially-available NAAT’s increasing, competition is making these highly sensitive tests more widely available than ever (See Methods chart, page 4-4 for details). Each of the different methods is based on similar principles of building copies of target nucleic acid sequences using various enzymes and probes and detecting amplified products by several different techniques. Performance has been shown to be comparable among all of the assays. Most significant differences between NAAT methods lie in logistical issues and relative costs rather than performance.

	<u>Culture</u>	<u>(i) Non-Culture, Non-Amplified</u>		
		EIA	DFA	NAP
Test Names/ Manufacturers	"Isolation in Cell Culture" <u>Cells</u> – BioWhittaker, Inc.; ViroMed, Inc.; American Type Culture Collection (ATCC). <u>FA Reagents</u> – Trinity Biotech (Syva MicroTrak), Bio-Rad Laboratories (Kallestad), VWR Scientific Products (Bartels).	"Enzyme Immunoassay" <u>Lab-based:</u> Trinity Biotech (Syva MicroTrak II), Bio-Rad Laboratories (Kallestad), Abbott (IMX Select), Biomerieux (Vidas) <u>Point of Care/Rapid:</u> BioStar (OIA), others	"Direct Fluorescent Antibody" -Trinity Biotech - MicroTrak® II (Syva), -Bio-Rad Laboratories (Kallestad) -VWR Scientific Products (Bartels).	"Nucleic Acid Probe" Gen Probe Inc
Principle	Chlamydia organisms from specimens are grown in lab-cultured cells, and detected after 24-72 hrs w/ fluorescent monoclonal antibody. <i>Note: Requires viable organisms.</i>	Chlamydial proteins from specimens isolated and detected by binding with an anti-chlamydial antibody, linked to an enzyme that will produce a visible color reaction	Specimen is smeared onto a glass slide; dried and fixed. Fluorescent anti-chlamydial antibody binds to organisms, making them visible under a high- powered FA microscope.	Target chlamydial RNA sequence hybridizes to probe; detection molecule produces light reaction read in a luminometer
Performance ~ Sensitivity ~ Specificity	50-85% ~100%	50-80% 95-99%	50-75% 95-99%	50-75% 95-99%
Specimen Requirements	Swabs (cervical, urethral, rectal, conjunctival, pharyngeal), resp. washings, abdominal fluid, semen, Use chlamydia culture-specific swabs and media recommended by testing lab (2SP, M4/M5, etc.)	Female cervical swabs; some also approved for ocular and male urethral swabs <i>Use only the specific collection and transport materials supplied by the testing laboratory.</i>	Urogenital, rectal, pharyngeal swabs	Cervical, male urethral swabs
Handling	Optimally within 24 hours, MUST be refrigerated. Can be frozen at -70°C (contact lab), <i>Do not freeze at -20°C</i>	Most tests, up to 7 days 2-30°C	7 days, 2-30°C	7 days, 2-30°C
TAT (time to perform test)	2-3 days	15 minutes to several hours	as little as 30-45 minutes	several hours
Special Advantages	Virtual 100% specificity; only forensically acceptable method for sexual assault/abuse (though NAAT may become acceptable)	Viability of organisms not required, so handling requirements less stringent; Some tests are inexpensive with high throughput; others can be done as patient waits	Only non-culture method for rectal or pharyngeal specimens (though recommended from symptomatic patients only)	Available gonorrhea test from same swab.
Potential Disadvantages	Technically demanding, less available, expensive, sensitivity can be low and is easily compromised by sub-optimal specimen collection and handling (viability loss)	Low sensitivity and specificity; inappropriate in medico-legal cases; cannot test urine specimens (male or female) limited appropriate specimens; confirmation of positives recommended	Expensive, highly dependant on technician skill; not suited to high-volumes	limited specimens; confirmation of positives recommended
<i>Please refer to individual tests' product inserts or contact your laboratory for specific details on each laboratory method described above.</i>				

	NAP-Signal Amplification	Nucleic Acid Amplification Tests (NAAT)		
		PCR	TMA	SDA
Test Names/ Manufacturers	Digene CT/GC Hybrid Capture II	“Polymerase Chain Reaction” Roche Diagnostics (Amplicor Cobas)	“Transcription Mediated Amplification” Gen Probe Inc. (Aptima)	“Strand Displacement Amplification” BD (Probe Tec)
Principle	DNA from specimens binds to RNA probe; hybrids captured by specific antibodies; many copies of enzyme-bound ab binds to captured hybrids, produces an “amplified” signal, measured on a luminometer	chlamydial DNA is amplified by heating and cooling cycles in the presence of nucleotides, specific primers and DNA-building enzymes; large amounts of nucleic acid produced are detected by various methods “true PCR” with detection via enzyme-substrate color reaction	ribosomal RNA; with DNA intermediates; no thermal cycler	simultaneous amplification and detection, flourescin-labeled detector probe
Performance ~ Sensitivity ~ Specificity	Limited data available; Sens. >NAP, < NAAT Spec. ~ = NAP		90-96% 98-99.8%	
Specimen Requirements	Female cervical and male urethral swabs, cervical Thin Prep Pap specimens	Female cervical and male urethral swabs; male and female urine, symptomatic or asymptomatic patients (Roche Cobas approved for CT/GC testing from Thin Prep Pap specimens)		
Handling		7 days, 2-8°C for swabs and urine; Urine up to 24 hr at 18-25°C (-20 °C, 30 days)	up to 7 days, swabs 2-25°C, urines 2-8°C (or 24 hrs 15-27°C) Store longer if frozen, -70°C	up to 6 days, swabs 2-27°C, urines 2-8°C (or 2 days 15-27°C with UPP) Do NOT freeze.
TAT (time to perform test)	5-6 hours	several hours to overnight		
Special Advantages	Improved sensitivity over other non-NAAT tests without the high cost; less technically demanding than NAAT, combination GC assay	Highest available sensitivity; applicable to less-invasive urine specimens (broadening screening availability in non-traditional settings); combination GC assay available for most NAATs		
Potential Disadvantages	Not cleared for urine or other alternative specimens; promised high sensitivity not yet proven	Still relatively expensive and technically demanding; may encounter performance issues (contamination) in inexperienced labs; sensitivity can be affected by amplification-inhibiting substances in specimens, still questionable for medico-legal cases; testing too soon after anti-chlamydial therapy may yield false-positive results, not approved for non-genital sites; no confirmatory tests available, transport requirements too stringent for some settings, some tests (LCR, Cobas) not suited to high-volume testing, urine not as sensitive as swabs in females.		
<i>Please refer to individual tests’ product inserts or contact your laboratory for specific details on each laboratory method described above.</i>				

	<u>Culture</u>	<u>Non-Culture, Non-Amplified</u>					<u>Amplified</u>	
		<u>Antigen Detection</u>			<u>Nucleic Acid Based</u>			
		<u>EIA</u>	<u>EIA-POC</u>	<u>DFA</u>	<u>NAP</u>	<u>Hybrid Cap.</u>	<u>NAAT</u>	
Examples: Test Names/ Manufacturers	"Isolation in Cell Culture" Cells, reagents available from a variety of manufacturers.	"Enzyme Immunoassay" Trinity Biotech (Syva MicroTrak II), Bio-Rad Labs (Kallestad),	"Rapid" or "Point of Care" Tests BioStar (OIA), others	"Direct Fluorescent Antibody" Syva MicroTrak®II, Kallestad Pathfinder, Bartels	"Nucleic Acid Probe" GenProbe PACE & PACE 2	Digene CT/GC Hybrid Capture II	Roche Amplicor/ Cobas (PCR); BD Probe Tec (SDA); Gen Probe Aptima (TMA)	
Principle	CT organisms grown in lab-cultured cells; detected after 24-72 hrs w/ fluorescent antibody. <i>Requires viable organisms.</i>	CT proteins detected by binding with anti-chlamydial antibody, linked to an enzyme that will produce a color change reaction	Same as EIA; proteins bound to a membrane, read visually	Specimen is fixed onto slide; fluorescent anti-CT antibody binds to organisms, visible under microscope.	Target chlamydial RNA sequence hybridizes to probe; detection molecule produces light reaction	CT DNA binds to RNA probe; hybrids captured by antibodies; copies of ab bind to captured hybrids, produces "amplified" signal	CT DNA is amplified by heating/ cooling cycles w/ nucleotides, specific primers and DNA-building enzymes; high copy numbers detected by various methods	
Performance: ~ Sensitivity ~ Specificity	50-85% ~100%	50-80% 95-99%	50-70% 90-97%	50-70% 95-99%	50-75% 95-99%	Sens. >NAP, < NAAT Spec. ~ = NAP	90-96% 98-99%	
Specimen Requirements (Contact your lab for specifics)	Urogenital, rectal, pharyngeal, conjunctival swabs; resp. washings, fluids, semen, in supportive medium.	Female cervical swabs; some also approved for ocular and male urethral swabs	Cervical swabs; some also approved for ocular, male urethral and urine	Urogenital, rectal, pharyngeal swabs	Cervical, male urethral swabs	Female cervical and male urethral swabs, cervical Thin Prep Pap specimens	Female cervical and male urethral swabs; male and female urine, (Roche also Thin Prep specimens)	
Use only the specific collection and transport materials supplied by the testing laboratory.								
Handling (Contact your lab for specifics)	Optimally within 24 hours of collection; MUST refrigerate. Freeze at -70°C for longer storage.	Most tests, up to 7 days 2-30°C	POC test should be performed same-day, on-site	7 days, 2-30°C	7 days, 2-30°C		Variable; 3-30 days, 2-30°C (check with lab for individual test requirements.)	
TAT (time to perform test)	2-3 days	several hours	15-30 minutes	as little as 30-45 minutes	several hours	5-6 hours	several hours	
Special Advantages	~100% specific; only appropriate method for sexual assault/ abuse (NAAT may become acceptable)	Viability not required, less stringent handling; Some tests relatively inexpensive	Results can be available while the patient waits, increasing treatment.	Only non-culture method for rectal or oral/ pharyngeal specimens (symptomatic only)	Available gonorrhea test from same swab.	Higher sensitivity than non-NAAT at lower cost; less demanding than NAAT, GC combo	Highest available sensitivity; good performance w/urine specimens, combo GC assay	
Potential Disadvantages	Technically demanding, expensive, sensitivity can be low and easily compromised (viability loss)	Low sensitivity and specificity; limited appropriate specimens (no urine)	Low sensitivity and specificity; limited appropriate specimens; Expensive	Low sensitivity and specificity; expensive, highly dependant on technician skill	Low sensitivity and specificity, limited specimens (no urine)	Sensitivity still lower than NAAT; no urine	Expensive, technically demanding, potential contamination issues, inhibition, not FDA cleared for 'alternate' specimens	

INTERPRETATION OF LABORATORY RESULTS

DEFINITIONS

A. SENSITIVITY

The ability of a test to detect infection if it is present. Another way to understand sensitivity is the ability of a test to correctly classify infected individuals as positive. Still another way to understand sensitivity is the percent of positive test results in a hypothetical population, all of whom have the infection. A perfectly sensitive test would be positive 100% of the time in such a population. Thus, a highly sensitive test gives few false negatives. The Sensitivity of a test is usually expressed as the percent of existing positives detected by the test.

$$\text{Sensitivity} = \frac{\text{“True” Positive results}}{\text{True Positives} + \text{False Negatives (positives missed)}}$$

B. SPECIFICITY

The ability of a test to detect absence of infection if it is NOT present, in other words, to correctly identify uninfected individuals as negative. Still another way to understand specificity is the percent of test results that are negative in a hypothetical population, none of whom have the infection. Thus, a test with high specificity gives few false positives.

$$\text{Specificity} = \frac{\text{True Negatives}}{\text{True Negatives} + \text{False Positives}}$$

C. POPULATION PREVALENCE

The proportion of individuals in a population who have the infection at a specific point in time. “Population” refers to a group of individuals with shared characteristics; risk of infection can vary significantly among individuals in a given population. However, since sensitivity and specificity are defined based on hypothetical populations who have 100% and 0% prevalence of the infection (respectively), and since real population have prevalence between 0% and 100% the predictive value of a test changes depending on the sensitivity and specificity of the test and the pre-test likelihood (i.e., population prevalence) of infection.

D. PRE-TEST LIKLIHOOD

The likelihood, prior to testing, that a given individual has the infection. Pre-test likelihood is usually estimated based on known or predicted population prevalence for the population appropriate to the individual (e.g. age group), then adjusted up or down based on the individual’s risk factors and/or signs and symptoms.

E. PREDICTIVE VALUES

The probability that a given result reflects the true status of the patient. “Positive Predictive Value” (PPV, PVP) is the **probability that a positive result is a true positive**, and is dependent on the *specificity* of the test and the *prevalence of infection* in the population. “Negative Predictive Value” (NPV, PVN) is the **probability that a negative result is a true negative**, and depends on the *sensitivity* of the test and the *population prevalence*.

$$\text{PPV} = \frac{\text{True Positive Results}}{\text{True Positive Results} + \text{False Positives}}$$

$$\text{NPV} = \frac{\text{True Negative Results}}{\text{True Negative Results} + \text{False Negatives}}$$

ESTIMATING PREDICTIVE VALUES

When estimating predictive values, the terms “population prevalence” and “pre-test likelihood” are often used interchangeably. In fact, the terms refer to predictive value for a group, the latter for an individual.

		“True” status		
		+	-	
Test result	+	a	b	(a+b)
	-	c	d	
		(a+c)	(b+d)	

Although “Prevalence” is not actually known, epidemiologic data allows for expected numbers of “true” results to be estimated.

False-positives = **b** False-negatives = **c**

Sensitivity	a/(a+c)	ability of the test to identify infected people
Specificity	d/(d+b)	ability of the test to correctly identify uninfected people
PPV	a(a+b)	probability of being infected if the test is +
NPV	d(c+d)	probability of actually being uninfected if the test is –

- Since the true prevalence is not known, predictive value estimates can be based on positivity/risk.
- PPV and NPV can be used to estimate the potential for false positive/negative results in a population and to assess the likelihood that an individual result accurately reflects a patient’s infection status.

Effect of Prevalence on Predictive Values:

Positive Predictive Value (PPV) is dependent on the prevalence of infection in the population being tested. PPV is highest where prevalence is high and is reduced in low-prevalence settings. Based on surveillance data, we know that in both urban and rural clinics prevalence is higher in patients meeting selective screening criteria (SSC) than in those not meeting SSC. The numbers below illustrate how differences in prevalence impact PPV and are based on a test with a sensitivity of 96.8% and specificity of 99.5%.

Non-Urban clinic, patients meeting SSC, Prevalence: 4.3%

		“True” status		
		+	-	
Test result	+	42	5	47
	-	1	952	
		43	957	1000

PPV: $a/(a+b) = 42/47 = \mathbf{89.4\%}$
NPV: $d/(c+d) = 952/953 = \mathbf{99.7\%}$

Non-Urban clinic, patients NOT meeting SSC, Prevalence: 1.1%

		“True” status		
		+	-	
Test result	+	11	5	16
	-	0	984	
		11	989	1000

$= 11/16 = \mathbf{68.8\%}$
 $= 984/984 = \mathbf{\sim 100\%}$

- The lower the prevalence (positivity, risk), the lower the PPV for any assay with a specificity of less than 100%.
- Because of the much higher proportion of negative results seen, differences in prevalence typically do not have as much impact on NPV.

SUMMARY: LABORATORY METHODS FOR CHLAMYDIA

Though a wide range of tests have been described, several important facts are true of all laboratory methods for CT:

1. Each test has specific collection materials that must be used; always use the exact materials supplied or recommended by the laboratory for a particular test. Failure to do so may compromise results or lead to specimen rejection by the laboratory.
2. Each test has specific handling requirements including acceptable temperature and time between collection and testing; again, follow specific lab recommendations for specimen handling. Failure to do so may compromise results or lead to specimen rejection by the laboratory.
3. All swab specimens must contain columnar epithelial cells for maximum sensitivity for any CT method used. Excess mucus, exudate, pus, blood and fecal material should be avoided.
4. The sensitivity and specificity of an assay are determined by comparisons with results of other assays and clinical information in controlled studies. Accuracy of sensitivity and specificity estimates are highly dependent on the standards of comparison used. Performance of laboratory assays in “real life” can vary from lab to lab, among different populations, and over time.
5. No laboratory test for chlamydia is 100% sensitive, and none, with the exception of culture, are 100% specific. False-positive and false-negative results can and do occur. It is impossible to determine the exact proportion of positives that are “false”; however, the likelihood of such results can be estimated in order to assist with interpretation of individual results. Always consider the *performance of the test* (sensitivity, specificity) and the *risk of infection* (population prevalence) when interpreting any individual test result. Remember that confidence in the accuracy of *positive* results is highest when testing patients at highest risk of infection.

CASE 2

One of the nurses in your clinic is concerned because she dropped the swab on the floor after the CT specimen was collected.

Questions

Should the specimen be used for testing? Why or why not?

**** To check your responses, see Case Study Discussions at the end of this section.**

CASE 3

You receive a phone call from the laboratory telling you that the swab used for the specimen was not from the appropriate collection kit.

Questions

Will this affect the test result? Why or why not?

** To check your responses, see Case Study Discussions at the end of this section.

CASE 4

A 26-year-old female presents to your clinic because she was recently treated for CT and wishes to make sure that she is cured.

Questions

Which CT tests are appropriate to use in this situation? Are there any special considerations with performing “test of cure” for CT?

** To check your responses, see Case Study Discussions at the end of this section.

CASE 7

A CT swab collected late Friday was put in the freezer to be mailed out Monday morning.

Questions

Can this specimen be tested?

** To check your responses, see Case Study Discussions at the end of this section.

CASE STUDY DISCUSSIONS

Case 1: Culture is the test of choice in cases of suspected sexual abuse or assault. Non-culture methods may produce false-positives and are not generally considered “forensically acceptable,” especially in cases involving children. In some cases a NAAT may be done in addition to culture for patient management purposes. NAAT may also be used in settings where culture is not available; however, results should be interpreted carefully.

Case 2: Obviously, you wouldn’t want to test that specimen! Environmental contaminants can interfere with lab tests in a variety of ways. If the patient has not left the clinic premises, it may be possible to substitute a urine specimen if collection of a replacement swab is not possible. Check with your laboratory to find out if female urine is acceptable for the test used in your facility.

Case 3: The laboratory should reject this specimen. The performance of lab methods can only be assured if the appropriate materials are used. Collection kits are as much a part of the test as the reagents or the instrumentation. In some cases certain materials such as wood-handled or calcium alginate swabs are known to interfere with results.

Case 4: Routine “test of cure” is not generally recommended for CT due to the effectiveness of treatment regimens. However, a re-screen 3-4 months after treatment of the initial infection may be prudent since risk behaviors often continue and re-infection rates are relatively high. In cases where a “test of cure” is done, culture is the preferred method due to its high specificity and the need for viable (infectious) organisms to produce a positive result. Non-culture methods may be used if sufficient time has elapsed for clearance of non-viable CT from the genital tract. In general, two weeks is considered sufficient for non-amplified tests and three weeks post-treatment if NAAT is used. As always, positive results should be interpreted with caution.

Case 5: The laboratory should reject this specimen. Laboratories are prevented by law from testing any specimen lacking a name or other “unique identifier” on the label.

Case 6: Columnar epithelial cells are the host cells for CT, which lives *inside* of cells. The presence of these cells is necessary for a specimen to be considered adequate. Excessive pus or mucus on the swab may indicate improper specimen collection technique and may interfere with the accuracy of test results.

Case 7: Handling conditions are determined by the particular assay in use. Freezing a specimen at regular freezer temperatures (usually -10 to -20 °C) may be contraindicated. Check with your lab for specific handling requirements. Improperly handled specimens may not be apparent to the lab, but they can yield unreliable results.

Case 8: An excess of blood can have an impact on the accuracy of results for some CT tests. Several options exist for testing this patient. If CT is strongly suspected to be causing this patient’s symptoms (or if other compelling risk factors are present), a cervical swab may be collected after carefully removing as much blood as possible, thus minimizing the amount of blood on the swab. Alternatively, urine may be submitted from female patients for some tests. Check with your lab.

Case 9: As long as the urine collected in the specimen container is the first part of the urine stream, the concentration of the actual urine is not an issue for chlamydia testing. Chlamydia, if present, will be washed from the walls of the urethra by the flow of urine, rather than being found in the urine or bladder itself.

DIAGNOSIS AND TREATMENT OF CHLAMYDIA

I. PATIENTS WHO SHOULD BE TREATED FOR CHLAMYDIA

A. Patients who have been screened and are found to be positive on any of the following commonly used tests for *Chlamydia trachomatis*:

1. EIA*
2. DNA probe*
3. Chlamydia culture*
4. Cervical/Urethral swab, urine PCR
5. Cervical/Urethral swab, urine SDA (ProbeTec)
6. Cervical/Urethral swab, urine TMA (GenProbe)
7. DFA*

* Sensitivity and/or specificity for this test are lower than for the other tests. If a patient is in a low risk epidemiologic group with a positive test, a physician should be consulted regarding management of this patient.

B. Patients who present with any of the following:

1. Contact with a sexual partner known to have chlamydia or gonorrhea
2. Urethral discharge with > 10 wbc per HPF
3. PID

C. All sexual partners exposed within 60 days prior to patient date of treatment.

D. Any person with gonorrhea, unless a simultaneous NAAT test for chlamydia was negative.

II. TREATMENT RECOMMENDATIONS

A. Non-pregnant Females/ Males

Recommended Regimens

- Azithromycin 1 gm po stat dose
- OR**
- Doxycycline 100 mg po bid X 7 days

Alternative Regimens

- Ofloxacin 300 mg po bid X 7 days
- OR**
- Erythromycin base 500 mg po qid X 7 days
- OR**
- Erythromycin ethylsuccinate 800 mg po qid X 7 days
-
- Levofloxacin 500 mg orally for 7 days

B. Pregnant Females

Recommended Regimens

- Erythromycin base 500 mg qid X 7 days
- OR**
- Amoxicillin 500 mg tid X 7 days
- OR**

Alternative Regimens

- Erythromycin base 250 mg po qid X 14 days
- OR**
- Erythromycin ethylsuccinate 800 mg po qid X 7 days
- OR**
- Erythromycin ethylsuccinate 400 mg po qid X 14 days
- OR**
- Azithromycin 1 gm po single dose

Contraindications

- Erythromycin estolate is contraindicated in pregnant women
- Doxycycline and other tetracycline-family antibiotics are contraindicated in pregnant women.
- Ofloxacin and other quinolone family antibiotics are contraindicated in pregnant women.

AMOXICILLIN

DOSAGE:

Amoxicillin 500 mg orally 3 times a day for 7 days

PREPARATION:

Capsules or tablets

ADVERSE REACTIONS:

As with other penicillins, it may be expected that untoward reactions will be essentially limited to sensitivity phenomena.

GI upset

CONTRAINDICATIONS:

A history of allergic reaction to any of the penicillins is a contraindication.

INTERACTIONS:

Chloramphenicol, erythromycins, sulfonamides, and tetracyclines may interfere with the bactericidal effects of penicillin. This has been documented *in vitro*; however, the clinical significance of this interaction is not well documented.

AGE RESTRICTIONS:

No restrictions

AZITHROMYCIN

DOSAGE:

Azithromycin 1 gm orally in a single dose

PREPARATION:

Powder or capsules

ADVERSE REACTIONS:

Anaphylaxis, GI upset

CONTRAINDICATIONS:

Allergy to any macrolide or any Enthromycin family antibiotics

INTERACTIONS:

H2 blockers and antacids may interfere with absorption, may increase theophylline levels and prolong PT in patients on Coumadin. This is unlikely to be a significant problem in patients taking single dose therapy.

PREGNANCY & LACTATION:

Category B considered safe in pregnant women if indicated

AGE RESTRICTIONS:

No restrictions

DOXYCYCLINE

DOSAGE:

Chlamydia and NGU

Doxycycline 100 mg orally 2 times a day for 7 days

PID/Epididymitis/Orchitis

Doxycycline 100 mg orally 2 times a day for 14 days

PREPARATION:

Capsules or tablets

ADVERSE REACTIONS:

Photosensitivity, GI upset, rash, anaphylaxis

PRECAUTIONS & CONTRAINDICATIONS:

Allergy to doxycycline or any tetracycline-family antibiotics

INTERACTIONS:

Antacids interfere with absorption

PREGNANCY & LACTATION:

Category D (Known teratogen) Do not use in pregnant or lactating females.

AGE RESTRICTIONS:

Not indicated in patients < 8 years of age (due to permanent scarring of teeth).

ERYTHROMYCIN

DOSAGE:

Chlamydia and NGU

Erythromycin 500 mg orally 4 times a day for 14 days

PREPARATION:

Caplets

ADVERSE REACTIONS:

GI (cramping, nausea, vomiting, and diarrhea), rash, anaphylaxis

Some clinicians recommend, if the 500 mg dose causes intolerable GI side-effects, that the patient may take 250 mg four times daily but double the remaining duration. (i.e., still take all prescribed pills). It is unclear whether this regimen is equally effective.

CONTRAINDICATIONS & PRECAUTIONS:

History of allergy to erythromycin or other macrolide-family antibiotics, patients taking terfenadine (Seldane) or other long-acting antihistamines

Erythromycin estolate is contraindicated in pregnant women.

INTERACTIONS:

Theophylline, Digoxin, ergotamine (increase levels of these drugs), Seldane and other long acting antihistamines (Leads to prolonged QT intervals and may cause sudden death)

PREGNANCY & LACTATION:

Category B (no restrictions). Erythromycin estolate is contraindicated in pregnant women.

AGE RESTRICTIONS:

No restrictions

OFLOXACIN

DOSAGE:

Ofloxacin 300 mg orally twice a day for 7 days

PREPARATION:

Tablets

ADVERSE REACTIONS:

GI upset, hematologic abnormalities, rash, anaphylaxis

PRECAUTIONS AND CONTRAINDICATIONS:

Quinolone allergy

Ofloxacin and other quinolones are contraindicated in pregnant women.

INTERACTIONS:

Antacids may interfere with absorption

Cimetidine may interfere with elimination

May increase cyclosporin levels

May increase levels of Theophylline

May increase levels of Coumadin

With nonsteroidal anti-inflammatory drugs, may increase the risk of CNS stimulation and convulsive seizures

PREGNANCY & LACTATION:

Category C (Not recommended for use in pregnancy or in lactating women).

AGE RESTRICTIONS:

Should not be used in patients < 16 yrs. due to possible effects on developing bone and cartilage

DATA

DEFINITIONS

Core Data Elements

The core data elements include date of birth, age, sex, race, ethnicity, state, the patient's county of residence (or other appropriate geographic locator including city or zip code), date of specimen collection, health care provider identifier, health care provider type/system type, laboratory identification number, specimen site (anatomical site from which the lab specimen was collected or urine from patient), disease code, laboratory test type, laboratory test result and actual reasons for testing a particular patient (to measure adherence to screening criteria).

Data

Factual information, especially information organized for analysis.

Project Definition

Core data elements as outlined on the data collection request form. Performance standard requires 90% completion of the core data elements.

If core data elements are missing, provider should be contacted to provide the necessary information.

Chlamydia testing surveillance data are used to:

- Identify, treat and counsel infected clients;
- Ascertain prevalence in testing sites/populations;
- Determine effectiveness of infertility prevention programs;
- Establish and re-examine screening criteria;
- Provide a quality assurance indicator;
- Justify funding.

Data review should:

- Be used to create site quarterly reports provided by the state alliance. These reports should be useful to specimen submitters to monitor and improve their services. Information that should be included is (but is not limited to):

- Test requisition form data completion rates; positivity rates;
- Unsatisfactory test result rate; reasons for unsatisfactory results
- Demographic elements of clients tested;
- Percent of total for each demographic element.
- List totals of specimens meeting each of the screening criteria; particularly note number of specimens without screening criteria listed on their test requisitions.

STATE DATA COLLECTION FORMS

DATA COLLECTION REQUEST FORM

- Every specimen must have an accompanying Chlamydia/Gonorrhea Request Form. Bar code label on the Request Form must be straight for proper scanning.
- The following data are recommended by Region V on the laboratory specimen request forms:
 - unique specimen identification number
 - state where specimen was tested
 - encrypted client identification number
 - county where client resides
 - zip code of client's residence
 - client's birthday or age at date of visit
 - client's gender
 - client's self-reported race
 - client's self-reported ethnicity
 - type of facility where specimen was collected
 - name or code of facility where specimen was collected
 - name or code of laboratory where specimen was tested
 - specimen collection date
 - type of chlamydia screening test
 - type of gonorrhea screening test
 - site of specimen collection
 - reason for test
- States may also have special data requirements.

CLIENT EDUCATION AND PARTNER REFERRAL SERVICES

Chlamydia Counseling/Education for Clients and Their Partners

Clients with a presumptive diagnosis of *Chlamydia trachomatis* (CT) or a confirmed positive CT test should be provided with the following information to assist them in understanding CT, especially its treatment and prevention. At a minimum education about CT should include:

- Name of the disease
- Signs and symptoms of the disease
- How the disease is transmitted
- Incubation period
- Possibility of having asymptomatic disease for an extended period of time
- Complications of untreated chlamydia for women, men and babies

Discussion of Chlamydia Treatment Should Include:

- Name of the drug(s) being used in treatment
- Quantity and frequency of drug usage
- Probable efficacy of treatment
- Potential side effects
- Food, drugs, conditions (e.g., sunlight exposure or behaviors that should be avoided; alcohol is not contraindicated during therapy)
- What to do if side effects occur or symptoms develop or do not resolve
- Importance of abstinence until self and partners have completed treatment
- Importance of completing medication, not missing doses, not sharing medication with partners

Behavioral Risk Reduction/Prevention Should Be an Integral Component of Patient Counseling and Should Include:

- Assisting clients in identifying personal risks for contracting/transmitting CT, i.e. unprotected sex, multiple partners, having sex when drunk or high, etc.
- Assisting clients in developing realistic, personalized risk reduction plans, i.e. condom usage, monogamy, refraining from sex when drunk or high, etc.

Risk Reduction Includes Management of Sexual Partners

The interviewing of sexually transmitted disease (STD) clients and the notification and referral of their sex partners for appropriate assessment and treatment is a strategy that has been in place for over fifty years.

Sex partner elicitation is defined as:

- The confidential, voluntary, and non-coercive facilitation of the examination and treatment of all sex partners of a client diagnosed with CT.

Sexual partners should be notified if the partner's last exposure occurred within 60 days prior to date of client's treatment:

- In order to intervene in the spread of disease
- To prevent the development of complications in infected partners and the original client if (s)he is re-exposed to an infected partner
- Where partner(s) can go for care, for example, a clinic or health department. Providing a referral card or letter facilitates the referral process

Confidentiality

For the purpose of this manual, confidentiality is defined as the maintenance of security of any personal information related to a client and his/her sexual partners as well as any information contained in the client's medical record.

To maintain the integrity and credibility of the site and to facilitate cooperation from the client, clinicians should discuss confidentiality with each client. All clients should be informed of the following:

- Partner names will only be used for field searches and notification
- In no instance will partners be told:
 - the name or identity of the original client
 - the geographic location where the exposure took place
 - the date or period of exposure
 - the specific type of exposure

Five Key Points in the Partner Counseling Format for Clients Diagnosed with Chlamydia

1. Introduction

- clinician's name and role
- purpose of session
- discussion of confidentiality
- providing client with test results

2. Address Patient Concerns

- What questions can I answer for you at this time?

3. Disease Comprehension and Five Key Motivating Points for Partner Referral

- sexual transmission of CT
- asymptomatic nature of disease
- risk of reinfection
- complications and consequences
- increased risk of acquiring or transmitting other STDs

4. Sex Partner Elicitation

- first: obtain the number of sex partners during critical period (60 days prior to date of treatment)
- second: obtain names of sex partners
- third: obtain date of last exposure and frequency of sexual exposures with partner

5. Coaching the Client's Referral of Sex Partners and Conclusion

- assessing client's ability to effectively conduct a referral of her/his sexual partners
- assessing client's safety or risk for domestic violence
- enhancing client's ability to effectively conduct a sex partner referral

For information regarding Partner Services Training, please contact your State Health Department. Additional training may be available.

QUALITY ASSURANCE

Each state within Region V may have different procedures regarding chart audits. Please consult your state health department for further details.

CHART REVIEW

Each clinic should incorporate *Chlamydia trachomatis* (CT) components into their quality assurance audit. A review and follow-up of the two most current data reports and 25 charts (or 10%, whichever is less) will provide most of the information needed to ascertain if the CT testing, treatment, and referral is meeting clinic standards. This review should include the following elements:

- Documentation of risk assessment history and education
- Compliance with clinic screening criteria for testing
- Documentation of test result, follow-up, and treatment, all being completed in a timely manner
- Informed consent for tests and treatment obtained
- Appropriate treatment prescribed
- Documentation of treatment or referral for sex partner(s)

REVIEW OF SYSTEMS THROUGH REPORTS

On a quarterly basis, review and analyze test results which include positive, negative, unsatisfactory and inconclusive. Investigate the reasons for unsatisfactory and inconclusive test results and document recommended methods for improvement.

Review the quarterly data quality assurance reports indicating missing data elements on laboratory and behavioral questionnaire forms (sentinel sites). Identify problem areas and document recommended methods for improvement.

CHECK SHEET FOR CHLAMYDIA TESTING, TREATMENT AND PATIENT COUNSELING/EDUCATION

Specimen Collection

- New staff must be trained in specimen collection and adequacy prior to independently collecting specimens.

Periodically

- Time yourself while rotating the swab in the endocervical canal (swab should be in canal for the time period specified on the manufacturer insert).
- Watch the specimen collection video (See appendix).
- Have another experienced clinician observe your performance and give you feedback.

Ask yourself

- Do I know the protocol for use of culture and when it is appropriate to use?
- Do I always clean the exocervix?
- Do I always rotate the correct swab in the correct site?
- Do I check to see that the swab is in the tube and the cap securely fastened?
- Do I check to see that the name or unique identifier is on the transport tube and that it matches the laboratory requisition form?
- Do I verify the patient risk history? Is it documented? Do I check out the inconsistencies with the client (e.g. never had sex but has three children)?
- Do I test within the clinic's screening criteria? Do I know the criteria?
- Do I notify patients of infection within the time frame of my clinic's protocol?
- Do I adhere to the CDC STD Treatment Guidelines for client treatment?
- Do I take the time to explain the treatment plan to the client adequately? Do I encourage questions? Is the plan in writing for the client to take with her/him? Do I document the referral?
- Do I stress the importance of treating sex partners and offer referrals for their examination and treatment?
- Do I discuss risk reduction and other prevention measures which the client believes (s)he can implement to avoid acquiring a STD in the future?
- Do I complete the laboratory requisition form in readable script?
- Do I ensure the client of confidentiality regarding his/her records and partner referral process?
- **Remember: If you didn't document it, it didn't happen.**

STAFF TRAINING

All staff working as part of the CT prevention program should have documented CT training pertaining to their area of expertise. Refer to the appendix for staff checklist. Identified training needs resulting from the review of reports and chart audits may be met through the state CT program. Contact your state training representative for more information on training programs that are offered to clinic sites. Please contact your state health department for more information regarding training opportunities.

GLOSSARY OF TERMS

Accuracy

The extent to which a measurement is close to the true value.

Amplification Test

A test which replicates the genetic material (DNA or RNA) of a microorganism such as chlamydia from a few copies to millions within a few hours. These amplified (replicated) copies can then be detected, usually by photometry or fluorimetry.

Analytical Range

The range of accuracy of a test, e.g. the values (results) of a glucose blood level may range from 10 to 10,000 units. However, if test A used to detect glucose is only capable of detecting from 100 to 1,000 units, then the analytical range of this test is 100 to 1,000 units.

Antibiotic

A chemical substance capable of destroying microorganisms, specifically bacteria.

Antibody

A type of serum protein that is produced by the body in response to bits of foreign organisms (antigens). Antibodies assist the body in removing or destroying foreign antigens and their associated organisms.

Antigen

Foreign substances (usually bits of organisms) that often stimulate the body to produce antibodies. Such substances may also be used in the lab to detect antibodies in the blood stream.

Asymptomatic

A state where a person is infected with a disease but has no clinical symptoms.

APHL

The Association of State and Territorial Public Health Laboratory Directors. The national organization of public health laboratory directors and workers representing state and territorial health departments.

Azithromycin

An antibiotic used to treat chlamydial infections that can be given in a single dose.

Bacterium

Any small, one-celled (unicellular) microorganism. Bacteria vary in shape (morphologically), being spheric (cocci), rod-shaped (bacilli), spiral (spirochetes), or comma-shaped (vibrio).

Batch

A set of specimens (e.g., endocervical swabs) processed and tested during a single run (diagnostic test).

Cervical Motion Tenderness (CMT)

Moderate to severe tenderness elicited when the cervix is palpated or manipulated.

Cervicitis

Infection and/or inflammation of the cervix. Can be a sign of chlamydial infection.

Cervix

The narrow neck of the uterus, which extends into the vagina.

Chlamydia trachomatis

Chlamydia trachomatis is the bacterial agent which causes chlamydial infections, the most common sexually transmitted bacterial infection in the United States. While chlamydia is classified as bacteria, they share some properties of both bacteria and viruses, and they reproduce only inside of epithelial cells.

CLIA

Clinical Laboratory Improvement Act of 1967 (and amendments of 1988) which sets the guidelines for any clinical laboratory which tests materials obtained from human patients, e.g.; blood, tissue, swabs, etc. CLIA is administered through the U.S. Health Care Financing Administration (HCFA).

Clinical Laboratory

A laboratory in which tests directly related to the care of patients are performed. Such laboratories use material obtained from patients for test, as compared with research laboratories, where animal and other sources of test material are also used.

Clinical Laboratory Procedure

Analytical procedure (test) performed on any specimens (samples) taken from humans and used to diagnose disease or infection.

Collection Sites

Locations in the body from which a chlamydia specimen may be taken. These sites include: cervix, urethra, rectum, throat, conjunctiva (eye).

Confirmatory Test

A test which is used to confirm positive screening results in order to minimize false positive results thereby improving specificity. This test employs a different target molecule than the screening test, e.g., *C. trachomatis* enzyme immunoassay (EIA) typically detects specific lipopolysaccharide (LPS); while direct fluorescent antibody (DFA) test, used to confirm a positive EIA test, targets the major outer membrane (MOMP) of *C. trachomatis*. This method is preferred to using a supplemental test (see Supplemental Test).

Control

An artificial specimen with a known value (i.e. positive or negative) which is included in every test run in order to monitor the performance of the test. For example, if the positive control would test negative, it would invalidate the results of that particular test run and specimens would have to be re-tested.

CSTE

Council of State and Territorial Epidemiologists. This is the national organization of epidemiologists working in state health departments.

Culture

A laboratory test involving the cultivation of microorganisms or cells in a special growth medium.

Cutoff (CO)

A mathematically derived calculation in any given immunoassay which is used to determine which specimens are positive (reactive) or negative (non-reactive). Generally specimens with values above the CO are positive and those below are negative.

Detection Limit

The range (limits) of detection of any test methodology, e.g. a *C. trachomatis* amplification test (NAAT) needs only 1-10 organisms to be present in order to detect CT, whereas an enzyme immunoassay (EIA) needs 100,000 organisms to be present in order to detect CT.

Diagnostic Test

A test designed to detect chlamydia in a patient presenting with symptoms or risk history, as distinguished from a screening test.

Direct Fluorescent Antibody Test (DFA)

The direct detection of chlamydia (antigen) from a specimen (e.g., endocervical swab, etc.) which is placed on a microscope slide and stained using fluorescently labeled chlamydia specific antibody. After proper staining, the slide is viewed under a fluorescence microscope. Chlamydia-positive specimens show apple-green elementary bodies in contrast to red background of counter-stained cells.

DIS (Disease Intervention Specialist)

A trained individual working with patients testing positive and their partners to confirm treatment and identify all other potentially infected individuals. Usually employed by a state or local health department.

DNA Probe

See Nucleic Acid Hybridization Test.

Doxycycline

An antibiotic used to treat chlamydial infections. The standard dosage for treatment of chlamydia is 100 milligrams twice a day, for 7 days.

Ectopic Pregnancy

A pregnancy occurring anywhere except in the uterus, usually in the fallopian tubes. A serious, potentially fatal consequence of chlamydial infection.

Ectopia

Visible columnar epithelial cells that extend onto the outer surface of the cervix. In younger women or women using hormonal contraceptives, ectopy is considered normal. However, ectopy increases the risk of acquiring chlamydia by exposing the more vulnerable columnar epithelial cells.

Enzyme Immunoassay (EIA)

A laboratory test that detects specific antigens or antibodies rather than the organism (e.g., chlamydia) itself.

Erythromycin

An antibiotic used to treat chlamydial infection, especially for pregnant women. The standard dosage is 500 mg orally 4 times a day for 7 days.

Etiologic Agent

An agent that causes disease.

External Quality Control

An external control (see control) specimen which is generally shared between multiple laboratories and the results compared for quality control purposes.

False-Negative (Result)

A test result that indicates the absence of a condition when the condition is actually present. The rate of occurrence of false-negative results varies with the diagnostic accuracy and the specificity of the test or procedure and the pre-test likelihood of disease. As the accuracy and specificity of a test increase, the rate of false negatives decreases. As the pre-test likelihood of disease increases the false negative rate also increases (except when sensitivity is 100%). Certain tests are known to yield false-negative results at a certain rate; in all tests, a small number of false negatives will occur by chance alone.

False-Negative (Rate)

The rate of occurrence of negative test results in subjects known to have the disease or behavior for which the individual is being tested. The rate of occurrence of false-negative results varies with the diagnostic accuracy and the specificity of the test or procedure and the pre-test likelihood of disease. As the accuracy and specificity of a test increase, the rate of false negatives decreases. As the pre-test likelihood of disease increases the false negative rate also increases (except when sensitivity is 100%). Certain tests are known to yield false-negative results at a certain rate; in all tests, a small number of false negatives will occur by chance alone.

False-Positive (Result)

A test result that wrongly indicates the presence of a condition when the condition is not present.

False-Positive (Rate)

The rate of occurrence of positive test results in tests of individuals known to be free of a disease or disorder for which the individual is being tested. The rate of occurrence of false-positive results varies with the diagnostic accuracy and the specificity of the test or procedure and the pre-test likelihood of disease. As the accuracy and specificity of a test increase, the rate of false positives decreases. Certain tests are known to yield false-positive results at a certain rate; in all tests, a small number of false positives will occur by chance alone.

Friability

Fragile, easily crumbled, especially prone to bleeding; for example, cervical tissue in some chlamydial infections.

Gonorrhea

A common sexually transmitted disease most often affecting the genitourinary tract and, occasionally, the pharynx, conjunctiva, or rectum. Infection results from contact with an infected person or by contact with secretions containing the causative bacteria *Neisseria gonorrhoea*.

Gray zone (GZ)

An artificially established range (zone) below a diagnostic test's cut-off (CO) value. The GZ generally ranges from 30-70% below the CO. Specimens in the established GZ are then re-tested by another methodology in order to increase the test sensitivity, i.e., to detect additional positive specimens.

Immunoassay

An assay (test) which detects antigens or antibodies.

Infertility

The inability to conceive or carry a fetus to term. Chlamydia-related infertility is most often caused by scarring in the fallopian tubes.

Inhibitor

A substance that interferes with the test's ability to detect the presence or absence of disease. Blood and mucus are examples of potential inhibitors for chlamydia.

Internal Quality Control

An internal control specimen made up and used by a particular laboratory (see control).

Kit

A package of test reagents, package insert, etc. which enables a laboratory to perform a particular test, e.g., a chlamydia kit would enable a laboratory to test for chlamydia.

LPS

The lipopolysaccharide in the chlamydia cell membrane, a part of the organism. The same LPS is present in all chlamydia species, e.g., *C. trachomatis*, *C. psittaci*, *C. pneumoniae*, etc. Any test which detects chlamydia LPS would cross react with all chlamydia organisms.

Lot

Diagnostic kits are manufactured in large quantities (lots). As part of quality control, laboratories record all results from each kit and lot in order to monitor for any variations which may occur among lots.

Mean

The numerical average of the results obtained from a series of analyses.

MOMP

The Major Outer Membrane Protein on the chlamydia organism. The MOMP is species-specific, i.e., *C. trachomatis* is different from *C. psittaci*, etc. Any test which detects MOMP will only react with each separate species, i.e., antibody against *C. trachomatis* MOMP will not react with *C. psittaci*.

Mucopurulent

Green or yellow discharge when viewed on a white cotton swab that has been inserted into the cervical os.

Nucleic Acid Hybridization Test (DNA Probe)

The Gen-Probe Pace 2 assay. A laboratory test which detects *C. trachomatis* ribosomal RNA.

OPA

Office of Population Affairs. This is the federal office which administers the Title X Family Planning Program and is part of the Department of Health and Human Services.

Package Insert (Test Kit)

The written pamphlet in every diagnostic test kit which includes instructions for proper use (kit directions) of the kit. In addition, the package insert contains some or all of the following: information on intended use; summary and explanation of the test; principles of the procedure; reagents provided; special precautions; specimen collection, storage and transport; materials provided (not provided with kit; procedural limitations; performance characteristics; results; and quality control.

Package Insert (Medication)

The written pamphlet for each prescription drug listing side effects, dosages, indications, contraindications, drug interactions, etc.

Partner Notification

The process of identifying sex partners of patients testing positive and informing them that they are at risk for infection and need to be tested.

Pelvic Inflammatory Disease (PID)

A clinical syndrome identified by a range of symptoms including lower abdominal pain and tenderness, bilateral adnexal tenderness, low-grade fever, and cervical motion tenderness. Serious sequelae (consequences) can include infertility, ectopic pregnancy, and chronic pelvic pain. PID can be one of the serious consequences of chlamydial infections.

Polymerase Chain Reaction (PCR)

DNA amplification test for chlamydia.

Predictive Value (Negative)

The likelihood that a person with a negative test does not have the disease, or in a group, the proportion of all negative tests that actually reflect a disease-free state.

Predictive Value (Positive)

The likelihood that an individual with a positive test has the disease, or in a group the proportion of all positive tests that actually reflect a disease state.

Presumptive Treatment

Also known as epidemiologic treatment. The treatment of patients suspected of having a disease based on identified risk factors and/or clinical findings without the confirmation of a test result.

Prevalence

The percentage of people in a given population that have a given disease, e.g., the prevalence of chlamydia in Clinic A is 5% that is 5 out of 100 individuals in Clinic A are infected with chlamydia.

ProbeTec

An amplification test for chlamydia. A process whereby a strand of DNA can be cloned (replicated) millions of times within a few hours by strand displacement amplification (SDA).

Proficiency Testing: (PT)

A program (CAP, AAB, etc.) in which samples (artificial patient specimens) are sent to participating laboratories for analysis. The true value (results) of the samples are unknown by the testing laboratory. The results are reported to the specific program (CAP, AAB, etc.) tabulated, compared to all participating laboratories and reported to the enrolling laboratory. PT specimens are an indicator of laboratory performance.

Qualitative

A test that is qualitative determines the presence or absence of a substance (antibody/antigen), e.g., an EIA detects the presence or absence of chlamydia.

Quantitative

A test that is quantitative determines the amount of a substance per unit volume or unit weight, e.g., blood glucose normal range 70-115 mg/dl-milligrams per deciliter.

Quality Assurance Program (QAP)

A comprehensive set of policies, procedures, and practices used to monitor the services provided in a clinical or laboratory setting. These plans should include protocols for proper record keeping, calibration and maintenance of equipment, monitoring of quality controls and proficiency testing results, and training.

Quality Control (QC)

The set of laboratory or clinical procedures designed to ensure that a test is working properly, e.g., test controls, monitor lot-to-lot variation, monitor/run CO values, etc.

Reagent

A substance that produces a chemical reaction in a sample that allows an analyte (the substance being measured) to be detected and measured.

Screening: Criteria

A set of characteristics used to determine which patients in an asymptomatic population should receive a test for chlamydia.

Screening Test

A test performed to detect chlamydia in a patient presenting for a routine exam, with no symptoms or risk history indicating chlamydia, as distinguished from a diagnostic test.

Selective Screening

Testing for chlamydia in a population using screening criteria, as opposed to universal screening of an entire patient population, or diagnostic testing of patients with symptoms.

Sensitivity

The ability of a test to accurately detect patients who have the disease or condition for which they are being tested.

Specificity

The ability of a test to accurately identify patients who do not have the disease or condition for which they are being tested.

Specimen

A small sample of something, intended to show the nature of the whole, e.g., a blood or urine specimen.

Specimen Adequacy

The quality of the specimen obtained from the patient judged by the number and type of cells sampled, e.g., in chlamydia testing, an endocervical specimen which contains any endocervical columnar epithelial cells.

Strand Displacement Amplification (SDA)

A DNA amplification method.

Supplemental Test

A test which is used to confirm positive screening results. This test employs the same target molecule as the original screening test, e.g., *C. trachomatis* enzyme immunoassays (EIA) typically detect specific lipopolysaccharide (LPS); the EIA blocking or neutralization assay also target this same molecule (LPS). As a general rule, results obtained from using one test should be confirmed using an alternate technology (see Confirmatory Test) in order to best decrease the incidence of false positive test results thereby increasing specificity.

Symptomatic

Presenting with clinical signs or symptoms of disease.

Title X

The federal legislation which supports federally funded family planning clinics.

Transcription Mediated Amplification (TMA)

RNA amplification test for the detection of chlamydia.

Turnaround Time (TAT)

The amount of time it takes to produce a test result from the time a specimen is received in the laboratory until it is reported out.

Universal Screening

Testing for chlamydia in an entire patient population, regardless of symptoms, risk history, or other factors.

Urethritis

Inflammation of the urethra.

INFERTILITY KNOWLEDGE ASSESSMENT PRE AND POST TEST

- 1. The incubation period for *Chlamydia trachomatis* (CT) is:**
 - a. 2-4 days
 - b. 7-21 days
 - c. 28-42 days

- 2. A female client is having a pap test, gonorrhea (GC) culture, and CT test done. The specimen for CT should be obtained:**
 - a. first
 - b. second
 - c. third
 - d. it doesn't matter

- 3. A specimen collection error that can affect CT test results is:**
 - a. presence of blood on swab
 - b. absence of mucous on swab
 - c. presence of epithelial cells on swab

- 4. Which of the following is true concerning the correct handling of male urine specimens for CT testing using Syva EIA?**
 - a. specimen will remain stable for up to 7 days if refrigerated
 - b. specimen should be shipped at ambient temperatures
 - c. specimen should be frozen if not shipped within 24 hours

- 5. Which of the following medications used for the treatment of CT are NOT contraindicated during pregnancy?**
 - a. azithromycin
 - b. doxycycline
 - c. erythromycin estolate

6. List 2 of the six minimum points to be included in education about CT (not including information about treatment).

1. _____
2. _____

7. Correct preparation of state data collection forms for CT testing includes:

- a. Under reason for test, mark “signs/symptoms” if purulent discharge is observed.
- b. Remove bar code from the request form and place in the client’s chart.
- c. Include the date of specimen collection on the request form.

8. Which of the following women would be considered most at risk for CT?

- a. A 17-year-old woman who has been with her new partner for the past 2 months.
- b. A 23-year-old woman in a mutually monogamous relationship who has an IUD.
- c. A 25-year-old single woman who uses a diaphragm for contraception.

9. Which of the following statements about salpingitis and pelvic inflammatory disease is true?

- a. Most women who have tubal infertility have had previously diagnosed salpingitis.
- b. After two episodes of PID, the rate of infertility increases.
- c. *Chlamydia trachomatis* infection in women is usually symptomatic.

10. Syva EIA and Gen-Probe can be used for collection of specimens from cervical, male and female urethral, rectal, and pharyngeal sites.

- a. True
- b. False

INFERTILITY KNOWLEDGE ASSESSMENT PRE AND POST TEST ANSWERS

1. B
2. C
3. A
4. A
5. A
6. * name of the disease
* signs and symptoms of the disease
* how the disease is transmitted
* incubation period
* possibility of having asymptomatic disease for an extended period of time
* complications of untreated CT for women, men and babies
7. C
8. A
9. B
10. B

DOCUMENTATION OF EMPLOYEE SUCCESSFUL COMPLETION OF RVIPP TRAINING MANUAL

This manual provides a synopsis of the history and background of the RVIPP and information regarding all components of the *Chlamydia trachomatis* (CT) prevention program. Each new employee of an RVIPP authorized CT screening site should complete the training manual within **30 days** of the first date of employment.

The following checklist should be reviewed for each new employee by the employee's supervisor. The supervisor should ensure that the employee has the skills to successfully accomplish items on the checklist which pertain to his/her responsibilities.

Employee:

- Provided copy of training manual
- Completed all sections of the manual, including Case Studies
- Understands state specific Family Planning or STD screening criteria
- Successfully completed a lab report form (form which accompanies specimen to laboratory)
- Successfully completed STD report form (form sent to state STD program documenting chlamydia infection)
- Examined specimen collection kit, including a "dry" walk through
- Reviewed specimen collection and genital exam tape
- Observed specimen collection and genital exam
- Understands agency policy regarding:
 - appropriate medications and when to use
 - guidelines for presumptive treatment
 - guidelines for informing client of positive test results
 - guidelines for partner referral and treatment

A score of **75%** should be achieved by the employee on the RVIPP training manual post test. The employee's supervisor should grade the post test. Both the employee and supervisor should sign in the designated areas below indicating successful completion of the manual. If the employee does not achieve 75% on the post test, technical assistance should be provided by the agency for the training manual modules in which the employee displayed deficiencies. If the agency cannot provide the technical assistance, the state's RVIPP designated training committee member should be contacted. A list of training committee members is provided on the final page of this manual.

The following statement should be completed for each new employee and submitted to the RVIPP designated training committee member for the appropriate state.

DOCUMENTATION OF EMPLOYEE SUCCESSFUL COMPLETION OF RVIPP TRAINING MANUAL

Name of Agency _____

Date of Employment _____ **Date of RVIPP training manual completion** _____

Score _____

Employee's signature _____

Supervisor's signature _____

Region V Infertility Prevention Project Self-Study Manual

Evaluation Form

Learning Objectives

Using a scale of 1 (not effective) through 5 (highly effective), please fill in the number next to how effectively this self-teaching manual enabled you to meet each of the following objectives.

	NOT EFFECTIVE		HIGHLY EFFECTIVE		
	①	②	③	④	⑤
1. Discuss the epidemiology and natural history of <i>Chlamydia trachomatis</i> (CT).					
2. Identify the clinical presentation and diagnosis.					
3. Identify your state's screening criteria.					
4. Describe specimen collection techniques.					
5. Understand the basic principles, utility and interpretation of commonly used laboratory tests for diagnosis of chlamydial infection.					
6. Teach elements and process of data collection.					
7. Effectively analyze and evaluate aggregate client demographics.					
8. Identify the components of a quality assurance program for CT screening in the clinic.					
9. Identify essential information that must be presented to the client.					
10. Identify strategies to facilitate behavior change.					
11. Describe treatment recommendation for CT and appropriate regimen based on patient profile.					
12. Rate the effectiveness of the self-teaching module.					
13. Were the objectives related to the overall purpose?	<input type="radio"/> yes <input type="radio"/> no				
14. How long did it take you to complete this activity?	_____ hours _____ minutes				

Nursing Continuing Education Application Form Instructions

Read each section of this module, and print a copy of the evaluation and the Post-Test.

Complete the post-test and the evaluation form. Make a copy of the completed forms for your records.

Mail a copy of the completed evaluation and post-test to HCET at the address below along with a check for the \$10 processing fee. Your nursing license number must be placed on the form in order for HCET to process the contact hour record. Checks should be issued and mailed to:

Health Care Education & Training, Inc.
9460 North Augusta Drive, #421
Carmel, Indiana 46032
Attention: Joyce Alley RNC

Your post-test will be graded. If a passing score of 75% is achieved, a contact hour record and post-test answer sheet will be forwarded to you in the mail. There are 2.4 contact hours available for the activity. Please forward any questions to Joyce Alley RNC at (317) 247-9008 or at JBalley@hcet.org.

Health Care Education & Training, Inc. (HCET) has been approved as a provider of continuing education in nursing by the Indiana State Nurses Association (ISNA). ISNA is accredited as an approver of continuing education in nursing by the Commission on Accreditation of the American Nurses Credentialing Center. The contact hours are recognized by all agencies affiliated with ANCC.

Nursing Continuing Education Application

I have enclosed a check or money order for \$10 payable to HCET. Payment must be included with your post-test and evaluation form to be processed.

LPN yes RN yes Nurse Practitioner yes

Nursing License Number

Name (please print)

Street Address

City

State

Zip

Current Job Title

() _____

Day Time Telephone Number

I certify that I completed the Infertility Prevention Self-Study Manual and I am eligible for Continuing Education contact hours.

Signature

Date

EDUCATIONAL RESOURCES

Videos

Sexually Transmitted Diseases in a Family Planning Setting, videotape, Cicatelli, 1999.
Overview of STD services in a family planning clinic.

An Approach to Taking a Sexual History in the 90's, videotape, Health Care Education & Training, Inc. 1997.

Video from a satellite workshop presented by Michael Policar, MD to help clinic staff develop interviewing skills related to sexual behaviors and relationships. Information obtained can be used with clients in planning or preventing pregnancy, preventing HIV and other STDs, and in identifying barriers to health such as substance use or threat of violence, for which referrals can be offered.

www.information@your.fingertips, video, Cincinnati STD/HIV Prevention Training Center, 1998. Designed to help healthcare professionals understand steps necessary to use the world wide web to access current information on the prevention, diagnosis and treatment of sexually transmitted diseases.

Web Links

Reproductive Health

American College of Obstetricians and Gynecologists
www.acog.com

Afraid to Ask?

www.afraidtoask.com

STD, reproductive health and general health information

Nursing Continuing Education Units (CEUs)

www.nurseceu.com/womn.htm

Health Care Education & Training, Inc (HCET)

<http://www.hcet.org>

Training curricula that provide contact hours for nurses and listings of training events and educational resources in Region V. Includes current information on CT and GC testing in Region V

Health Square: Women's Health & Prescriptive Drug Resource

www.healthsquare.com/drugmain.htm

National Association of Nurse Practitioners in Women's Health
www.npwh.org

Planned Parenthood Federation of America
www.plannedparenthood.org/

General Health

Alan Guttmacher Institute
www.agi-usa.org

National Library of Medicine's PUBMED
www.nlm.nih.gov/
Free access to Medline

The Kaiser Family Foundation
<http://www.kff.org/>

STDs

American Social Health Association
www.ashastd.org
Source for pamphlets and patient information on STDs

Center for Disease Control & Prevention (CDC)
www.cdc.gov
Entry to publications, health topics, research, guidelines for all CDC programs and services

CDC En Espanol
www.cdc.gov/spanish/
Provides health related information the Hispanic/Latino professional and to the Spanish speaking/oriented community in general

CDC Prevention Guidelines
<http://aepo-xdv-www.epo.cdc.gov/wonder/PrevGuid/prevguid.html>

Cincinnati STD/HIV Prevention & Training Center
<http://www.stdptc.uc.edu/default.cfm>
Calendar of educational courses for health care professionals offered through classroom and distance learning. Site contains an image library

Sexually Transmitted Diseases Treatment Guideline 2002
<http://www.cdc.gov/mmwr/preview/mmwrhtml/rr5106a1.htm>

Sexuality Information & Education Council in US (Siecus)
www.siecus.org

HIV/AIDS

Centers for AIDS Prevention Studies-UCFS

<http://www.caps.ucsf.edu/>

CDC Division of HIV/AIDS Prevention

<http://www.cdc.gov/hiv/dhap.htm>

HIV/AIDS Insite

<http://hivinsite.ucsf.edu/InSite>

Add 2-letter state abbreviation for resources and information on HIV/AIDS in any state

HIV/AIDS Information Center

www.medscape.com

Midwest AIDS Training & Education Centre (MATEC)

<http://www.uic.edu/depts/matec/>

National Institutes of Health

<http://www.aidsinfo.nih.gov/>

The Body

<http://www.thebody.com/>

UNAIDS

<http://www.unaids.org/en/default.asp>

Publications

Centers for Disease Control & Prevention

Morbidity & Mortality Weekly Report (MMWR)

Sexually Transmitted Diseases Treatment Guideline 2002

Available online at <http://www.cdc.gov/mmwr>

Or order through

Superintendent of Documents

US Government Printing Office

Washington DC 20402

Chlamydia Care Quality Improvement Toolbox

California Chlamydia Action Coalition

Tulip Graphics, Inc

2920 Seventh Street

Berkley, CA 94710

(510) 878-0000

available for download at:

http://www.ucsf/castd/downloadable/clinicalpractice_guidelines.pdf

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