Neisseria gonorrhoeae
by Real-time PCR with Reflex to Antibiotic resistance by Molecular Analysis

“The sensitivity and specificity of the nucleic acid amplification tests (NAATs) are clearly the highest of any of the test platforms for the diagnosis of chlamydial and gonococcal infections. Since accurate diagnosis is the goal, there is no justification for the ongoing use of other technologies”(1, 2).

- Centers for Disease Control and Prevention (CDC)

- MDL provides detection of Neisseria gonorrhoeae by Real-Time PCR, one of the most powerful and sensitive gene analysis techniques available.
- Sensitivity and specificity up to 99%.
- Test results are typically available within 24-48 hours.
- This test has been validated for detection of N. gonorrhoeae using the OneSwab®, UroSwab® (males and females), and ThinPrep®.

Epidemiology

- Gonorrhea is the second most commonly reported bacterial STD in the United States with an estimated 700,000 new N. gonorrhoeae infections occurring each year (7).
- Due to the fact that gonococcal infections among women are frequently asymptomatic, targeted screening of young women at increased risk for infection is a primary component of gonorrhea control in the United States (7).

Pathogenesis

- Neisseria gonorrhoeae, a Gram-negative diplococci, is the causative agent of gonorrhea.
- Due to its affinity for columnar or pseudo stratified epithelium, it is most commonly detected in the genital tract with the primary site of involvement being the endocervical canal and transition zone of the cervix.
- N. gonorrhoeae’s unique ability to alter surface structures allows increased pathogenicity, facilitates epithelial surface attachment, and enables evasion of the host’s immune response.
- Transmission of N. gonorrhoeae occurs almost exclusively through sexual contact, though it can also be transmitted via the passage of a neonate through an infected mother’s birth canal or via autoinoculation from the hands of an infected person to their eye.
- Incubation time for this infection is typically 3-5 days and transmission more frequently occurs from male to female.
- Some risks factors for infection include: low socioeconomic status, early onset of sexual activity, unmarried status, a history of previous gonorrhea infection, illicit drug abuse, and prostitution.

Clinical Significance

- Clinical manifestation in men usually includes symptomatic urethritis; however, pharyngeal, anorectal, and disseminated infections are also possible.
- In women, infections are often asymptomatic; however, when manifested, symptoms may include: vaginal discharge, dysuria, intermenstrual bleeding, menorrhagia, pelvic discomfort, infection of the periurethral glands, Bartholin glands, and anorectum.
- Due to the fact that gonorrhea can have serious consequences for both mother and neonate, it is crucial to screen pregnant women for infection who reportedly have an incidence of gonorrhea during pregnancy as high as 10%.
- Complications that can occur during pregnancy include: amniotic infection syndrome, premature rupture of the membranes, chorioamnionitis, premature birth, intrauterine growth retardation, neonatal sepsis, and postpartum endometritis.
- During vaginal delivery with an infected mother, 30% to 35% of neonates will acquire Neisseria gonorrhoeae which, if left untreated, can progress to corneal ulceration and scarring, as well as blindness called gonorrheal ophthalmia neonatorum.

Laboratory Diagnosis

- Diagnosis of infections with N. gonorrhoeae has traditionally relied upon Gram stain, culture, and immunochemical techniques. Although culture techniques may be highly specific, sensitivity is greatly impacted by the adequacy of the clinical specimen and transport conditions, particularly when transporting to off-site facilities.
- Due to the genome plasticity of N. gonorrhoeae strains circulating in the population, this bacterium has developed resistance to multiple classes of antimicrobial agents, resulting in decreased efficacy for gonorrhea therapy. An increase of ceftriaxone-resistant N. gonorrhoeae demonstrated a similar pattern to previous reports in Japan and Southeast Asia that prompted the CDC to remove ciprofloxacin from the treatment guidelines as a primary antibiotic (1-3).
- In August 2012, the CDC called for Ceph-R NG surveillance through N. gonorrhoeae antibiotic susceptibility testing for patients that have failed treatment (3). Although susceptibility testing by culture remains the standard for antibiotic susceptibility determination in clinical microbiology, there are inherent growth-related issues that can delay results by as much as three days or more.
- Known mechanisms of antibiotic resistance in N. gonorrhoeae are linked to mutations in the chromosomal DNA as well as the presence of plasmid-borne genes. Surveillance of genetic markers of antibiotic resistance is important for the prediction of clinical resistance as the antibiotic susceptibility signatures of individual N. gonorrhoeae strains differ.

NOTE: PenR = penicillinase producing Neisseria gonorrhoeae and chromosomally mediated penicillin-resistant N. gonorrhoeae; TetR = chromosomally and plasmid mediated tetracycline-resistant N. gonorrhoeae, and QNG = quinolone-resistant N. gonorrhoeae.

**Figure 1:** Gonococcal isolate surveillance project (GISP)-penicillin, tetracycline, and ciprofloxacin resistance among GISP isolates, 2010 (7).

- Test 167 Neisseria gonorrhoeae by Real-Time PCR (Reflex to Antibiotic Resistance by Molecular Analysis) developed by MDL, offers a valuable diagnostic tool for the reliable detection of genetic determinants of antibiotic resistance, thereby predicting antibiotic susceptibility of individual N. gonorrhoeae strains in a given clinical specimen. This test addresses the problem of genetic variability in N. gonorrhoeae and delivers a prognostic recommendation for antibiotic therapy in a personalized manner.
Table 1: Comparison of Multiple Assay Systems for the Detection of Neisseria gonorrhoeae.

<table>
<thead>
<tr>
<th>Test</th>
<th>N</th>
<th>Prevalence (%)</th>
<th>Sensitivity (%)</th>
<th>Specificity (%)</th>
<th>PPV (%)</th>
<th>NPV (%)</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>PCR</td>
<td>100</td>
<td>7.8</td>
<td>100</td>
<td>99.4</td>
<td>93.4</td>
<td>100</td>
<td>(17)</td>
</tr>
<tr>
<td>Amplicor</td>
<td>2238</td>
<td>5.2 *</td>
<td>96.3</td>
<td>98.7</td>
<td>80.2 *</td>
<td>99.8 *</td>
<td>(18)</td>
</tr>
<tr>
<td>Aptima Combo 2</td>
<td>1479</td>
<td>8.6</td>
<td>99.2</td>
<td>98.7</td>
<td>88.1</td>
<td>99.9</td>
<td>(19)</td>
</tr>
<tr>
<td>BD Probe Tec</td>
<td>1411</td>
<td>8.1</td>
<td>97.2</td>
<td>99.4</td>
<td>91.6</td>
<td>99.6</td>
<td>(20)</td>
</tr>
<tr>
<td>GEN-PROBE (Pace 2)</td>
<td>1750</td>
<td>8.7</td>
<td>97.1</td>
<td>99.1</td>
<td>90.6</td>
<td>99.8</td>
<td>(21)</td>
</tr>
<tr>
<td>Culture</td>
<td>866</td>
<td>4.5</td>
<td>50.0</td>
<td>97.1</td>
<td>40.0</td>
<td>98.0</td>
<td>(22)</td>
</tr>
</tbody>
</table>

† = Unless otherwise noted, all specimens are swabs
Ϯ = Calculated data

Screening

Table 2: Summary of screening for N. gonorrhoeae infection by nucleic acid amplification testing (NAAT) (derived from 7).

<table>
<thead>
<tr>
<th>Women</th>
</tr>
</thead>
<tbody>
<tr>
<td>Annual routine screening for all sexually active women at risk for infection.</td>
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<tr>
<td>Screening at the first prenatal visit for all pregnant women at risk or living in a high prevalence area.</td>
</tr>
<tr>
<td>In women with cervicitis via either vaginal, cervical, or urine samples.</td>
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</tbody>
</table>

<table>
<thead>
<tr>
<th>Men who have sex with men (MSM)</th>
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<tbody>
<tr>
<td>Screening for urethral infection via nucleic acid amplification testing (NAAT) of urine in all men who have had insertive intercourse the preceding year regardless of condom use.</td>
</tr>
<tr>
<td>Screening for rectal infection via nucleic acid amplification testing (NAAT) of a rectal swab in all men who have had receptive anal intercourse during the preceding year.</td>
</tr>
<tr>
<td>Screening for pharyngeal infection via nucleic acid amplification testing (NAAT) in all men who have had receptive oral intercourse during the preceding year.</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Both Men and Women</th>
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<tr>
<td>Newly diagnosed HIV infection</td>
</tr>
</tbody>
</table>

Treatment

Table 3: Current Recommendations from the CDC for adults, adolescents & children >45 kg: urogenital, rectal infection with N. gonorrhoeae (16).

**Recommended Regimens**

- Ceftriaxone 250 mg IM in a single dose **PLUS**
- Azithromycin 1 g orally in a single dose

OR

**Doxycycline** 100 mg orally twice a day for 7 days

**Alternative Regimens:** If ceftriaxone is not an option

- Cefixime 400 mg orally in a single dose **PLUS**
- Azithromycin 1 g orally in a single dose

OR

**Doxycycline** 100 mg orally twice a day for 7 days

Due to the concerns for developing patterns of antimicrobial resistance, most current recommendations for treatment should be followed. Guidance can be obtained from the CDC website (http://www.cdc.gov/std/gisp) and state and local health departments.

If treatment is still unsuccessful, contact the CDC for a consultation.


REFERENCES: