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Chlamydia trachomatis and Neisseria gonorrhoeae user information

At Micropathology Ltd. we offer PCR testing for *Chlamydia trachomatis* (Ct) and *Neisseria gonorrhoeae* (Ng).

Chlamydia trachomatis

Chlamydia trachomatis is a gram-negative, non-motile, obligate intracellular bacterium which exists outside host cells as inactive elementary bodies but can infect host cells via endocytosis and reside as membrane protected inclusion bodies intracellularly.

There are 19 known serovars and three biovars which cause distinct clinical syndromes:

- Serovars A, B, Ba and C cause chronic follicular keratoconjunctivitis or 'trachoma' which may lead to chronic scarring and is the most common cause of preventable blindness worldwide
- Lymphogranuloma venereum: LGV (serovars L1, L2, L2a and L3) causing an endemic STI in Africa, India, South-East Asia, South America and the Caribbean. More frequently detected in the UK in MSM (men who have sex with men) rectal swabs.
- Serovars D-K cause both common STI infections resulting in urethritis, pelvic inflammatory disease, ectopic pregnancy and infertility, but can also affect neonates born to positive mothers causing pneumonia and conjunctivitis.

Chlamydia is the most commonly diagnosed bacterial STI in the UK. As infection often has no symptoms but can have serious health consequences, the National Chlamydia Screening Programme (NCSP) recommends that Chlamydia screening should be offered as an integrated component of existing sexual and reproductive health services and through a range of other settings such as primary care and internet-based testing to ensure that young people (<25 years) have universal access to testing. From June 2021, NCSP opportunistic testing for the asymptomatic was refocused on women only.

A further consideration is neonatal chlamydial conjunctivitis, often within one to five days post birth after infection through the birth canal. Around 30-50% of neonates born to infected mothers will develop a conjunctivitis of which progression is usually rapid and serious complications such as visual impairment and perforation are not uncommon, even with prompt treatment, 6% of cases resulted in conjunctival scarring. Up to 20% of neonates exposed to chlamydial infection develop pneumonia.

Due to the fastidious nature of this intracellular organism and the requirement for a very high throughput method, NAATs are exclusively the test of choice for detection of *C. trachomatis*. The PCR often targets the cryptic plasmid due to its high copy number making these assays very sensitive. At Micropathology we target the cryptic plasmid in a probe-based PCR.

The 2015 BASHH Chlamydia trachomatis guidelines recommend preferred sample types of vulvo-vaginal swabs for women, and first-catch urine for men. Endocervical swabs in women and urethral swabs in men are also acceptable alternative sample types. Rectal swabs and pharyngeal swabs are the accepted extra-genital sample types. Eye swabs are also appropriate in cases of conjunctivitis.

The *C. trachomatis* assay at Micropathology is validated for testing of swabs, urine, semen, and thin preps (endocervical cellular specimens). Other sample types will be accepted for testing but will be reported with a caveat stating that the assay is validated for testing this sample type. This assay is currently awaiting UKAS accreditation.

Specimens sent for Ct or Ng are tested on both the Ct and Ng assays. Samples received for LGV DNA testing are first confirmed as containing *Chlamydia trachomatis* DNA using the Ct assay before being run on the LGV assay, following the 2013 BASHH LGV guidelines.

Neisseria gonorrhoeae

Neisseria gonorrhoeae is a gram-negative intracellular diplococcus that only infects humans and causes the sexually transmitted disease gonorrhoea. It is the second most common bacterial STI in the UK. This organism has become a major health concern due to association with poor reproductive outcomes and rising rates of antibiotic resistance.

Penile urethral infection

Over 90% of individuals are symptomatic, with the primary symptoms being urethral discharge and dysuria. Testicular and epididymal pain and swelling may also be seen but is less common.

Endocervical infection

Approximately 50% of infections are symptomatic. The primary symptom is increased or altered vaginal discharge. Gonorrhoea may also cause intermenstrual bleeding and menorrhagia, but these symptoms are rare. Pelvic and lower abdominal tenderness is observed in around 25% of cases, and may be an indication of *Chlamydia trachomatis* coinfection.

Rectal infection

Mostly asymptomatic, but symptoms can include anal discharge and perinala/anal pain. Up to a third of urogenital infections in women present with rectal infection.

Pharyngeal infection

Occasionally associated with sore throat but generally asymptomatic. Most commonly detected through screening and contact tracing.

Complicated infection

If the primary infection is not adequately treated, secondary complicated may occur. Ureathral or endocervical infections may cause epididymorchitis, prostatitis, or pelvic inflammatory disease (PID). PID is reported in approximately 14% of women with gonorrhoea. Haematogenous dissemination may also occur, causing skin lesions, arthralgia, arthritis, tenosynovitis, endocarditis, meningitis, or perihepatic inflammation.

As *N. gonorrhoeae* is always considered a pathogen, where it is detected, it should always be treated to prevent further complications and onward spread. A test of cure is recommended for all cases due to antibiotic resistance and the severity of further infection and contact tracing should be conducted to prevent continued transmission. Infection cannot be ruled out in individuals who test within two weeks of sexual contact with an infected partner, therefore BASHH guidelines recommended that patients return for repeat testing after this window period if treatment is not initially given. Additionally, as approximately 19% of patients with gonorrhoea have concurrent *C. trachomatis* infection, so testing for other STIs should be undertaken where *N. gonorrhoeae* is detected.

Multiple methods can be used for the diagnosis of gonorrhoea. For immediate results, microscopy can be used to identify intracellular gram-negative diplococci in swabs from males with urethral discharge. The primary role of culture is now for antimicrobial susceptibility testing, which is of increasing importance as antimicrobial resistance in *N. gonorrhoeae* continues to evolve and spread. All individuals with gonorrhoea diagnosed by NAAT should have cultures taken for susceptibility testing prior to treatment.

Use of NAAT tends to be more sensitive than culture, particularly in oropharyngeal and rectal sites, showing high sensitivity (>95%) in both symptomatic and asymptomatic infection. Many available *N. gonorrhoeae* NAAT assays have cross-reactivity with some closely related *Neisseria* species, therefore BASHH recommend confirmation of positives using a different assay, to achieve an overall positive predictive value over 90%. NAATs may also be crucial in diagnosing infection in extra-genital sites such as from joint fluid or heart valves.

The Micropathology assay is a nested PCR targeting the 16S rRNA gene of *Neisseria gonorrhoeae*.

The 2018 BASHH *Neisseria gonorrhoeae* guidelines recommend preferred sample types of vulvo-vaginal swabs for women, and first-catch urine for men. Endocervical swabs in women and urethral swabs in men are also acceptable alternative sample types. Rectal swabs and pharyngeal swabs are the accepted extra-genital sample types. These sample types are all UKAS accredited for testing on the *Neisseria gonorrhoeae* assay at Micropathology. Additionally, thin preps (endocervical cellular specimens), eye swabs, and semen are UKAS accredited for testing on the assay. Other sample types will be accepted for testing but will be reported with a caveat stating that the assay is not UKAS accredited for this sample type.

Specimens sent for Ct or Ng are tested on both the Ct and Ng assays. Any specimens deemed to contain *N. gonorrhoeae* DNA are sent for sequencing for additional confirmation.

References

Nwokolo, Nneka C., et al. "2015 UK national guideline for the management of infection with Chlamydia trachomatis." *International journal of STD & AIDS* 27.4 (2016): 251-267.

White, John, Nigel O'Farrell, and David Daniels. "2013 UK national guideline for the management of lymphogranuloma venereum: clinical effectiveness group of the British association for sexual health and HIV (CEG/BASHH) Guideline development group." *International journal of STD & AIDS* 24.8 (2013): 593-601.

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