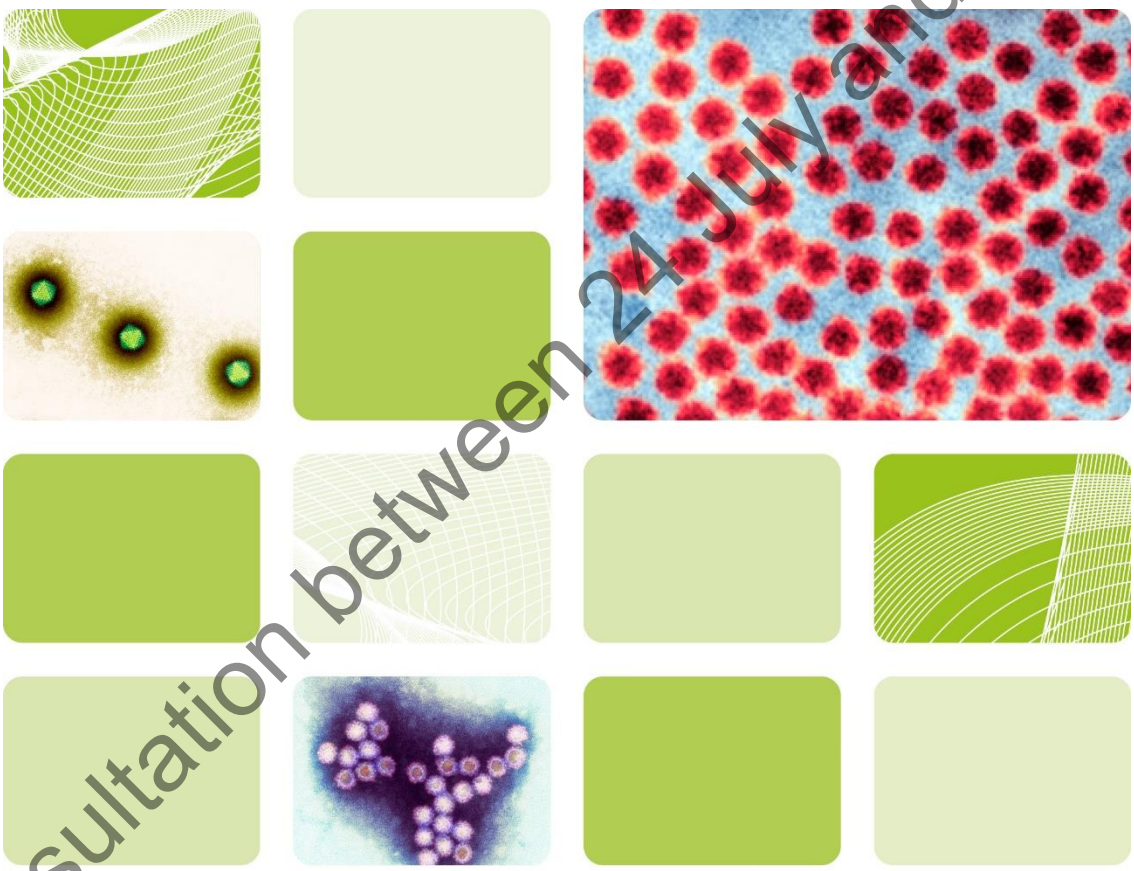
UK Health Security Agency

UK Standards for Microbiology Investigations

Chlamydia and Gonorrhoea infection – testing by Nucleic Acid Amplification Tests (NAATs)



National Institute for Health and Care Excellence (NICE) has renewed accreditation of the process used by the **UK Health Security Agency** to produce **UK Standards for Microbiology Investigations (UK SMIs)**. The renewed accreditation is valid until **30 June 2026** and applies to guidance produced using the processes described in '**UK Standards for Microbiology Investigations Development Process' (2021)**. The original accreditation term began on 1 July 2011.

Issued by the Standards Unit, Specialised Microbiology and Laboratories, UKHSA

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UK Standards for Microbiology Investigations (UK SMIs) are developed under the auspices of UKHSA working in partnership with the partner organisations whose logos are displayed below and listed on the UK SMI website. UK SMIs are developed, reviewed and revised by various working groups which are overseen by a steering committee.

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Contents

Amendment table

Each UK SMI document has an individual record of amendments. The amendments are listed on this page. The amendment history is available from standards@ukhsa.gov.uk.

Any alterations to this document should be controlled in accordance with the local document control process.

Amendment number/date	x/dd.mm.yy
Issue number discarded	
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Anticipated next review date*	dd.mm.yy
Section(s) involved	**Amendment**

*Reviews can be extended up to 5 years where appropriate

1 General information

View general information related to UK SMIs.

2 Scientific information

View scientific information related to UK SMIs.

3 Scope of document

This UK SMI covers the testing of clinical samples for the investigation of urogenital *Chlamydia trachomatis,* lymphogranuloma venereum (LGV) and *Neisseria gonorrhoeae* infection by Nucleic Acid Amplification Tests (NAATs).

This UK SMI does not cover ocular trachoma or neonatal infections (including pneumonia and conjunctivitis). The use of point of care tests (POCTs), where the test result can be delivered to the patient without the sample being sent to the laboratory, is not covered in this UK SMI. NAATs which are not approved for extra-genital and pooled specimens should be validated locally. Enzyme immunoassays (EIA) tests are not recommended (1).

Refer to UK SMI Q 4 - Good laboratory practice when performing molecular amplification assays.

This UK SMI should be used in conjunction with other UK SMIs.

Definitions

The following definitions apply:

During testing process

Reactive – Initial internal-stage positive result pending confirmation.

Not reactive – Initial internal-stage negative result.

Equivocal or Indeterminate – Result is not clearly positive or negative. Further testing is required.

Self-sampling – the individual collects their own specimens following instructions and send them back to the laboratory for testing

Pooled samples - individual specimens (swabs, urine or blood) are combined into a pooled specimen to reduce the cost of screening

Reporting stage

These terms are used for final or preliminary reports:

Detected – Report-stage confirmed reactive result.

Not detected – Report-stage not reactive result.

Indeterminate – Reactive result that cannot be confirmed.

Inhibitory – The presence of inhibitors within the sample has prevented amplification. A further specimen is required. The term used may be different for various platforms, for example “invalid”.

Reporting of invalid/inhibitory results should be based on manufacturer’s interpretation.

4 Introduction

Chlamydia

Chlamydia is caused by the bacterium *Chlamydia trachomatis* and is the most common sexually transmitted infection (STI) in the UK. It is most common in young people aged 15-24 years. Spread is primarily via sexual transmission however newborn babies can acquire chlamydia infection from the birth canal during delivery which can present as conjunctivitis or pneumonia in the neonate.

Most cases are asymptomatic; however the patient can have the following signs and symptoms:

- Persons with a vagina: vaginal discharge, intermenstrual bleeding, dysuria, lower abdominal pain, dyspareunia, mucopurulent cervicitis, pelvic tenderness and cervical motion tenderness.
- Persons with a penis: urethral discharge and dysuria.

Extra genital infections can also occur such as rectal infection and pharyngeal infection.

If chlamydia is not treated, it can lead to other complications such as pelvic inflammatory disease (PID), pregnancy outside the womb (ectopic pregnancy) infertility, endometritis, salpingitis, sexually acquired reactive arthritis, perihepatitis and long term pelvic or abdominal pain (1).

Gonorrhoea

Gonorrhoea is caused by the Gram-negative diplococcus *Neisseria gonorrhoeae.* The key communities that have a disproportionate burden of gonorrhoea are young people aged 15-24 years, gay, bisexual and other men who have sex with men (GBMSM), people of the black Caribbean ethnic community and people living in the most deprived areas. It is spread by sexual contact through the vagina, anus and by oral sex (2). Refer to UK SMI ID 6: Identification of Neisseria species for more information.

Gonorrhoea rates increased 7.5% from 79,268 diagnoses in 2022 to 85,223 diagnoses in England (3). Antimicrobial resistance to first and second line therapeutics is increasing (4). The British Association of Sexual Health and HIV (BASHH) recommend ceftriaxone as first line therapy. Ceftriaxone resistance is most common in the Asia- Pacific region and is occasionally detected in the UK in people who have travelled to or moved from this region (5).

Lymphogranuloma venereum (LGV)

Lymphogranuloma venereum (LGV) is a sexually transmitted infection caused by 3 serovars of the bacterium *C. trachomatis:* serovars L1, L2 and L3. Symptoms can be complex, severe and may involve multiple sites in the body such as the genitals, the anus, rectum, oral cavity and lymph nodes (6,7). The incubation period can range from 3 – 30 days from the time of contact with an infected individual. Patients may present with proctitis however asymptomatic infection may occur (7). There have also been increases in LGV cases from 1,173 cases in 2022 to 2,069 in 2023. These are usually less frequently reported (3).

LGV testing from rectal swabs is recommended for patients with proctitis, or from GBMSM, with or without symptoms, with *C. trachomatis* infection at any site (1,7).

4.1 Screening

The National Chlamydia Screening Programme (NCSP) has been developed to focus on reducing the harms from untreated chlamydia infection. The harmful effects of chlamydia occur predominantly in women and persons with a womb or ovaries (this includes transgender men, and non-binary people assigned female at birth, and intersex people with a womb or ovaries) under the age of 25. The NCSP recommends that they are offered a chlamydia test (8,9).

Chlamydia screening in community settings, such as GPs and pharmacies, will only be proactively offered to young women and persons with a womb or ovaries. Everyone can still get tested if they need, but men and persons with a penis will not be proactively offered a test unless an indication has been identified, such as being a partner of someone with chlamydia or having symptoms (9). Services provided by sexual health services remain unchanged.

Testing for gonorrhoea is recommended in any setting where it is clinically indicated, such as for symptomatic patients, contacts of those infected or attendees of sexual health clinics (10).

5 Safety considerations

The section covers specific safety considerations (11-30) related to this UK SMI, should be read in conjunction with the general safety considerations.

C. trachomatis and *N. gonorrhoeae* are hazard group 2 organisms. Refer to current guidance on the safe handling of all sample types and organisms documented in this UK SMI.

The above guidance should be supplemented with local COSHH and risk assessments.

6 Public health management

For information regarding notification refer to:

https://www.gov.uk/government/collections/notifications-of-infectious-diseases-noids

For further information on public health management refer: https://www.gov.uk/government/collections/chlamydia-surveillance-data-screening-and-management.

For information regarding the National Chlamydia Screening programme refer to:

https://www.gov.uk/government/collections/national-chlamydia-screening-programme-ncsp

Also refer to the BASHH guidelines and NICE guidelines for the management of *Chlamydia trachomatis* infection and lymphogranuloma venereum (7):

https://www.bashh.org/current-guidelines/all-guidelines/

https://cks.nice.org.uk/topics/chlamydia-uncomplicated-genital/

Partner notification should be discussed at the time of diagnosis. All sexual partners of patients with a positive *C. trachomatis* and *N. gonorrhoeae* NAAT should be offered full STI screening (1,31).

7 Specimen processing and procedure

7.1 Specimen type

Specimen types: Urine, vulvo-vaginal swabs, urethral swabs, endocervical swabs, rectal swabs, oropharyngeal swabs (self-collected or clinician-collected).

Note: Urine in women should be avoided.

7.1.1 Laboratory diagnosis

Compared to culture, NAATs have shown to be most sensitive and specific for the detection of *C. trachomatis* and *N. gonorrhoeae.* It is therefore recommended in this UK SMI for diagnosis and screening. NAATs may also be used for the investigation of extra-genital infections where validated locally. In medico-legal cases a positive NAAT should be confirmed using a second NAAT with a different genomic target (1) and for specific patient groups/specimen types a second NAAT should be used to confirm *N. gonorrhoeae* positives. The majority of commercially available testing platforms are capable of detecting *C. trachomatis* and *N. gonorrhoeae* from a single specimen. Typing of LGV is performed by a separate test but in most cases will be able to use the original specimen which tested positive by chlamydia NAAT (32,33).

The sampling sites should relate to the type of sexual activity reported and the patient group (see Table 1) (1). The recommended sample type for persons with a vagina is a vulvovaginal swab (VVS) which may be self-collected.

Endocervical swabs have been shown to be less sensitive than vulvovaginal swabs when self-collected and must be taken by a healthcare worker (1). The testing of first

catch urine specimens from women/ persons with a vagina should only be used if other specimens are not available (1,33).

In men/ persons with a penis, first void urine has been shown to be more sensitive than urethral sampling and is the sample type of choice (1). Urine should be held for a minimum of one hour and the first 20mL sampled upon subsequent urination.

Rectal samples may be taken during proctoscopy, or directly by the patient or healthcare worker. Local validation should be carried out for testing extra-genital specimens. Refer to UK SMI Q 1 Commercial and in-house diagnostic tests: evaluations and validations for further information.

Throat swabs may also be taken by clinician or self-collected.

In all cases laboratories should follow manufacturers' instructions regarding individual specimen types.

Table 1. Appropriate sample sites for *C. trachomatis* and *N.* gonorrhoeae NAAT dependent on sexual activity (1)

	Type of sexual intercourse:		
Patient	**Oral**	**Vaginal**	**Anal**
GBMSM[a]	Receptive oro-anal: rectal swab Receptive: oropharyngeal swab Insertive: 1st void urine		Insertive peno-anal: 1st void urine Receptive peno-anal: rectal swab Receptive oro-anal: rectal swab
Heterosexual male/ persons with a penis	Insertive: 1st void urine	Insertive: 1st void urine	Insertive peno-anal: 1st void urine
Heterosexual female/ persons with a vagina	Receptive: oropharyngeal swab	Receptive: Vulvovaginal Swab	Receptive: rectal swab

Footnotes

a) In GBMSM, where there is also sexual activity with women, refer to heterosexual male for appropriate sample type following vaginal sexual activity.

7.2 Specimen transport and storage conditions

Samples should be stored in an appropriate transport media and transported to the laboratory within 24 hours of collection.

Please refer to the Guidance for the design of self-sampling packs and associated support for self-sampling processes within Sexually Transmitted Infection and Blood Borne Virus testing for more information.

7.3 Specific technical limitations

NAAT inhibition

It has been recognised that samples may contain different lubricants or substances that can cause inhibition, potentially causing false negative results (33). It is recommended to use an inhibition control in NAAT testing (1). Internal and cellular controls exist within commercial platforms. Many NAATs are able to remove inhibitory substances during the nucleic acid extraction process. Rectal specimens, urine from pregnant women and urine from women in the third week after menstrual bleeding may contain high levels of inhibitors (it is likely that hormones have a role to play in this inhibition) (33). In duplex or multiplex assays, where several targets may be detected, competitive inhibition may be observed. The test manufacturers' instructions should be followed as they may contain a list of substances which have been identified as inhibitory through verification and validation. The laboratory may consider regular monitoring of inhibition levels and positive rates.

Contamination

The risk of contamination should always be considered when using NAATs (34,35). See also UK SMI Q 4 Good laboratory practice when performing molecular amplification assays.

8 Investigation

8.1 Laboratory tests

Chlamydia trachomatis

NAATs are the recommended tests and are known to be more sensitive and specific than EIAs. NAATs have varying sensitivities and specificities for diagnostic tests in urogenital specimens have been demonstrated in clinical trial data, manufacturers' validation data and published papers.

If the sample is associated with a medico-legal case, then testing with a different genomic target is required even in a high risk population. NAATs should be performed on all sites where penetration has occurred (1).

Samples can be sent to the Sexually Transmitted Infections Reference laboratory (STIRL) for LGV testing, or a local laboratory with validated test. Acceptable sample

types include residual clinical specimens in which *C. trachomatis* has been detected by the local laboratory (by NAAT) or extracted DNA samples.

Neisseria gonorrhoeae

Microscopy of Gram stained specimens enables the direct visualisation of *N. gonorrhoeae* as Gram negative diplococci. Microscopy sensitivity is increased in person with a penis who have discharge and people with rectal symptoms. Microscopy is not recommended in specimens from people without symptoms, pharyngeal specimens and female urethral/cervical specimens due to reduced sensitivity.

Culture remains primarily for antibiotic susceptibility testing and to detect resistant *N. gonorrhoeae.* Resistant *N. gonorrhoeae* is a global concern as it continues to evolve and spread. A culture specimen should be taken from people with suspected or confirmed gonorrhoea infection prior to treatment. Culture sensitivity is increased when time from sample collection to plating is minimised thus direct plating in the clinic is advised or timely transfer to laboratory in transport media for immediate plating. Molecular methods to assess genomic sequences/regions which may confer resistance to antibiotics are becoming available.

NAATs have high sensitivity (>95%) in samples from patients with and without symptoms. NAATS are also recommended for pharyngeal and rectal specimen testing (locally validated) however it should be noted commercial NAATs may cross react with commensal *Neisseria* species especially in the pharynx. It is recommended to confirm positive results with a separate molecular target in samples from a population with a test positive predictive value of <90%, especially extra-genital specimens.

Refer to UK SMI ID 6: Identification of Neisseria species

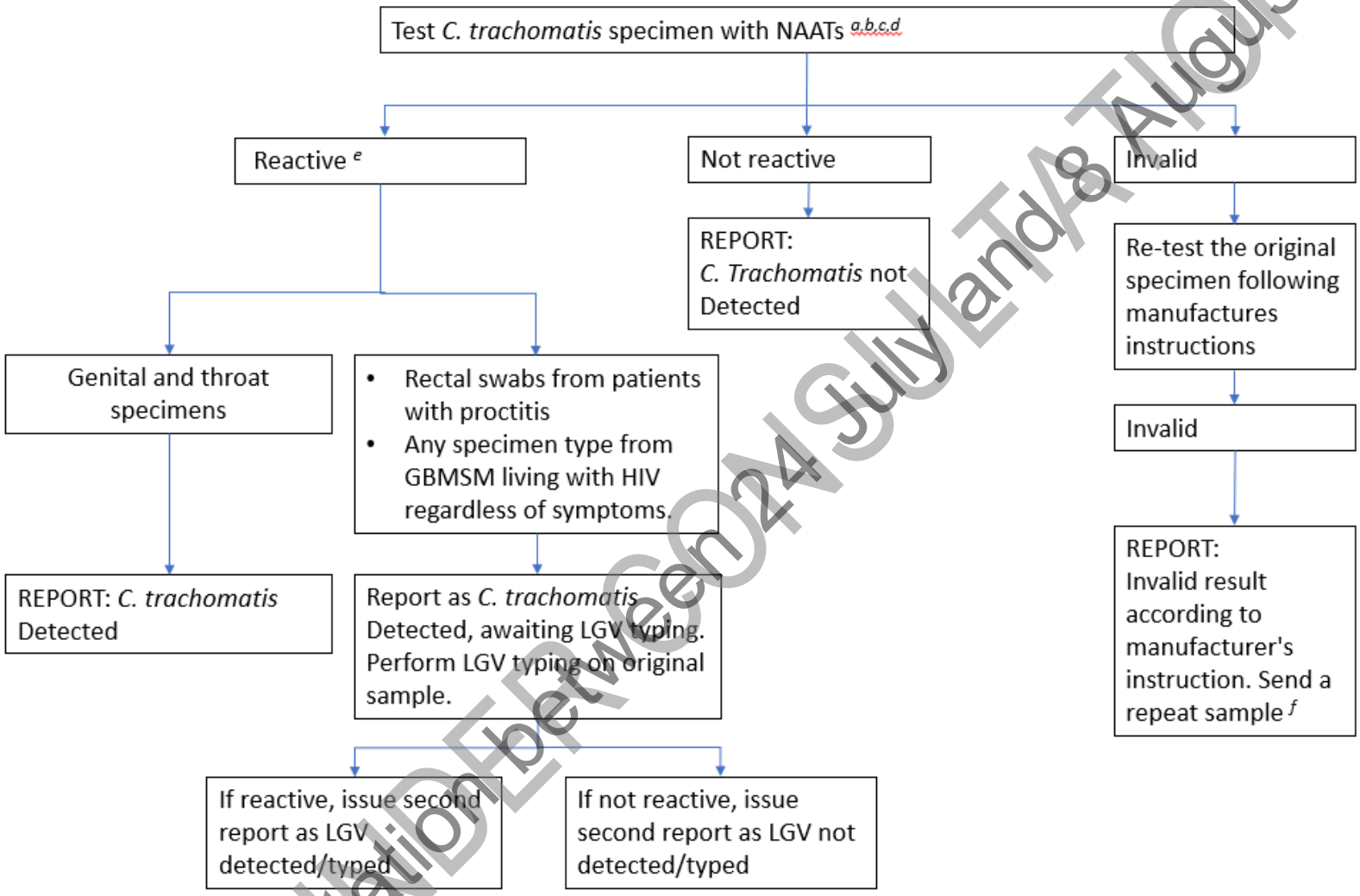
8.2 Window period and test of cure

Patients should be offered testing when they first present. If there has been possible sexual exposure within the previous two weeks, patients should return for a repeat test two weeks following exposure (1). Refer to BASHH guidelines for further information.

8.3 Medico-legal cases

Where results are likely to have medicolegal significance, specimens should be handled in accordance with Royal College of Pathologists' Guidance for handling medicolegal samples and preserving the chain of evidence and BASHH National Guideline on the management of STI and related conditions in children and young people. Legal precedent is limited but, for best practice, laboratories should confirm a reactive NAAT result by using a different target to ensure accurate results (1).

9 Algorithm 1: Testing for *Chlamydia trachomatis* infection



9. Footnotes

a) Recommended specimen types are first catch urine (preferred) or urethral swab for men/person with a penis and vulvovaginal swab (which may be self-collected) for women/ persons with a vagina.

b) Laboratories using dual NAAT capable of detecting both *C. trachomatis* and *N. gonorrhoeae* should follow nationally agreed algorithms and confirmatory strategy for the *N. gonorrhoeae* component of the test.

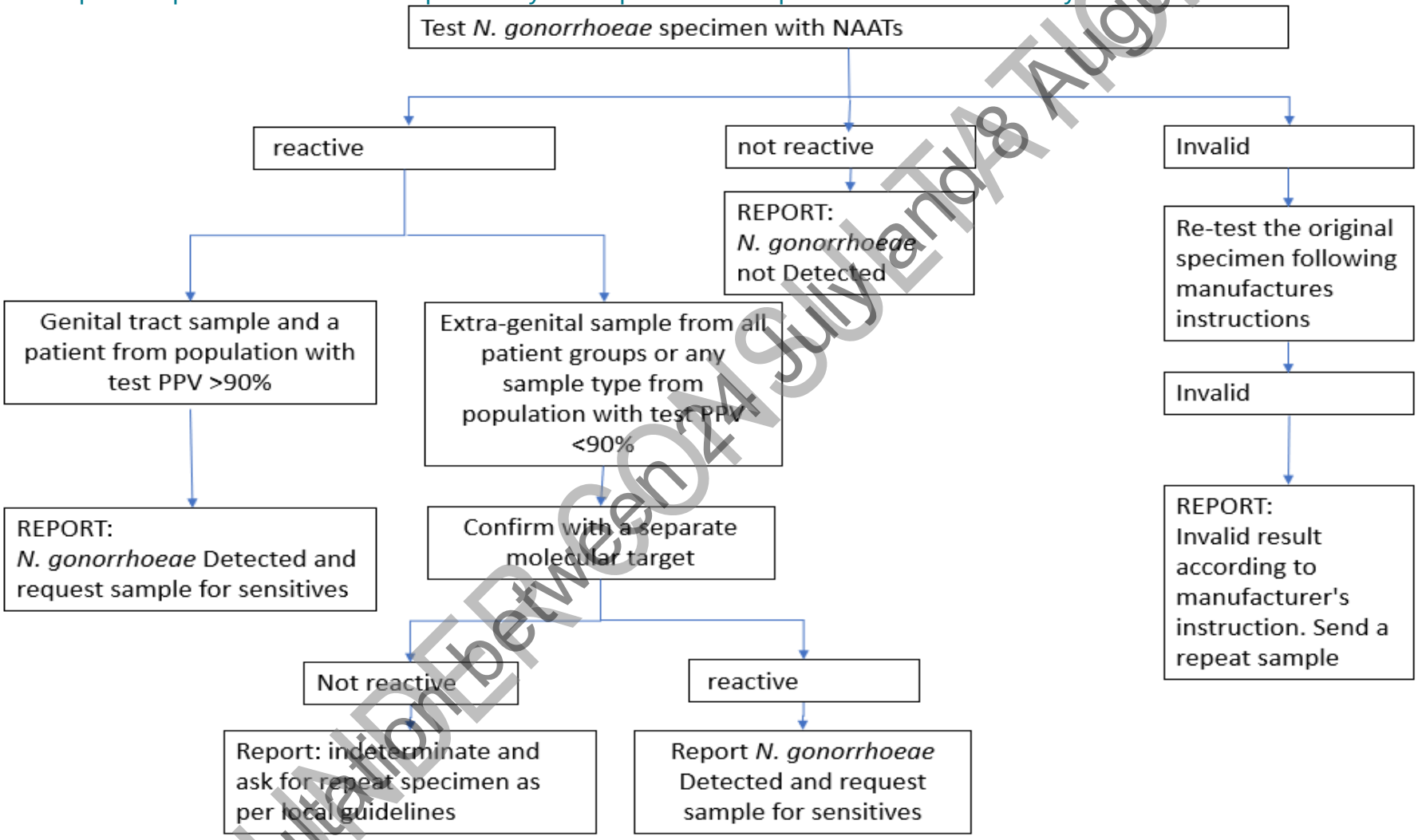
c) Laboratories should follow good practice when undertaking molecular testing. For *C. trachomatis* this should include environmental sampling. See UK SMI Q 2 Quality assurance in diagnostic virology and serology laboratory and UK SMI Q 4 Good laboratory practice when performing molecular amplification assays for further information.

d) It is recommended to use an inhibition control for each specimen (1). Failure to do so may lead to false negative results.

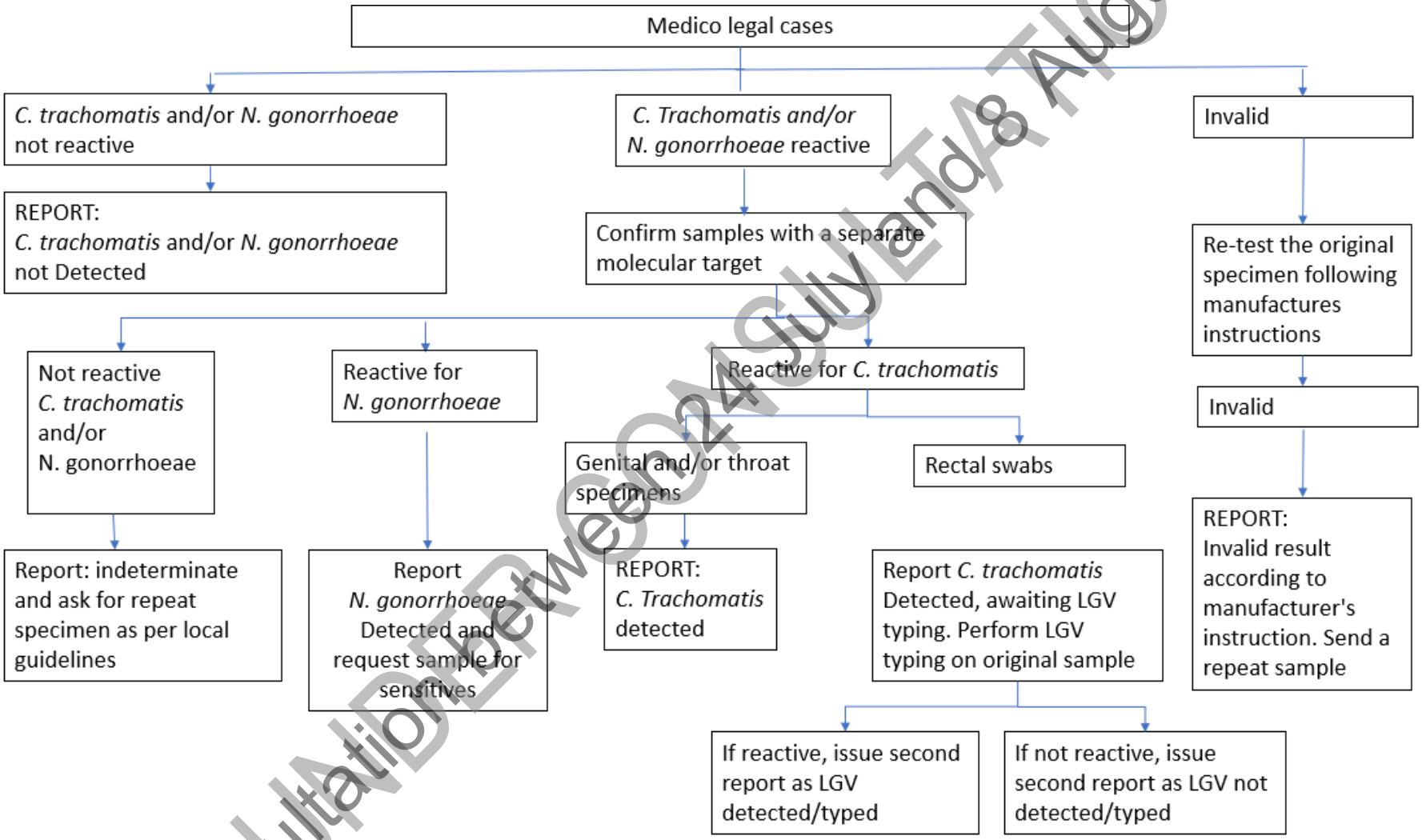
e) Many authorities no longer recommend testing with a second target unless testing is associated with a medico-legal case (1). The decision on whether to retest with a second platform depends on the sample type (for example samples from extra-genital sites such as rectal swabs), the platform that has been used for screening and the prevalence of *C. trachomatis* in the population tested.

9.1 Algorithm 2: Testing for *Neisseria gonorrhoeae*

PPV – positive predictive value is the probability that a person with a positive test result actually has the tested infection



9.2 Algorithm 3: Testing for Medico-legal cases



10 Interpreting and reporting laboratory results

***C. trachomatis* screening assay**	***C. trachomatis* confirmatory assay***	**Reported result**	**Interpretative comment**	**Notes**
Reactive	N/A	Detected	*C.trachomatis* DNA detected in sample	Refer relevant samples for LGV typing
Reactive	Reactive	Detected	*C. trachomatis* DNA detected in sample	Refer relevant samples for LGV typing
Not Reactive	N/A	Not Detected	*C. trachomatis* DNA not detected in sample	
Reactive	Not Reactive	Indeterminate	Initial reactivity has not confirmed. Consider sending repeat specimen.	

N. gonorrhoeae screening assay	***N. gonorrhoeae* confirmatory assay***	**Reported result**	**Interpretative comment**	**Notes**
Reactive	N/A	Detected	*N. gonorrhoeae* DNA detected in sample	
Reactive	Reactive	Detected	*N. gonorrhoeae* DNA detected in sample	
Not Reactive	N/A	Not Detected	N. gonorrhoeae DNA not detected in sample	
Reactive	Not Reactive	Indeterminate	Initial reactivity has not confirmed. Consider sending repeat specimen.	

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An explanation of the reference assessment used is available in the scientific information.

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