Gonorrhoea

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Toc blurb
Gonorrhoea is a sexually transmitted infection caused by the bacterium *Neisseria gonorrhoeae* that affects millions of people worldwide, and its incidence is increasing in many settings. The emergence and spread of antimicrobial resistance in *N. gonorrhoeae* threatens to leave affected individuals with no effective treatments.

Abstract
The bacterium *Neisseria gonorrhoeae* causes the sexually transmitted infection (STI) gonorrhoea, which has an estimated global annual incidence of 86.9 million adults. Gonorrhoea can present as urethritis in men, cervicitis or urethritis in women, and in extragenital sites (pharynx, rectum, conjunctiva, and rarely systemically) in both sexes. Confirmation of diagnosis requires microscopy of Gram-stained samples, bacterial culture or nucleic acid amplification tests (NAATs). As no gonococcal vaccine is available, prevention relies on promoting safe sexual behaviours and reducing STI-associated stigma, which hinders timely diagnosis and treatment thereby increasing transmission. Single-dose systemic therapy (usually injectable ceftriaxone plus azithromycin) is the recommended first-line treatment. However, a major public health concern globally is that *N. gonorrhoeae* is evolving high levels of antimicrobial resistance (AMR), which threatens the efficacy of the available gonorrhoea treatments. Improved global surveillance of the emergence, evolution, fitness and geographical and temporal spread of AMR in *N. gonorrhoeae*, and improved understanding of the pharmacokinetics/pharmacodynamics for current and future antimicrobials in the treatment of urogenital and extragenital gonorrhoea is essential to inform treatment guidelines. Key priorities for gonorrhoea control include strengthening prevention, early diagnosis, and treatment of patients and their partners; decreasing the stigma; expanding surveillance of AMR and treatment failures; and promoting responsible antimicrobial use and stewardship. To achieve these goals, the development of rapid and affordable point-of-care diagnostic tests that can simultaneously detect AMR, novel therapeutic antimicrobials and especially gonococcal vaccine(s) is crucial.

[H1] Introduction

The sexually transmitted infection (STI) gonorrhoea remains a major public health concern globally. The aetiological agent of gonorrhoea, the bacterium *Neisseria gonorrhoeae* (the gonococcus), generally causes mucosal infections of the urogenital tract, predominantly infecting columnar and transitional epithelium, although it can also attach to the stratified squamous epithelium of the ectocervix[1,2]; such *N. gonorrhoeae* infections most frequently result in urethritis in men and cervicitis in women, but urethritis in women is also observed)[3,4]. This obligate human host-adapted pathogen was described for the first time by Albert Neisser in Gram-stained microscopy of urethral discharge in 1879 (Ref[5]). *N. gonorrhoeae* is a diplococcal (that is, it is typically composed of two joined cells with the adjacent sides flattened, resulting in a characteristic kidney or coffee bean appearance in microscopy), Gram-negative microorganism; it belongs to the bacterial class *Betaproteobacteria* and the family *Neisseriaceae*, and has been co-evolving with its human host for centuries. The family

Neisseriaceae comprises the genus *Neisseria* and other genera such as *Kingella* and *Eikenella*[6-8]. *Neisseria* genus currently consists of at least 23 species, of which about half are human-restricted species, some are animal-restricted and some can be isolated from mucosal surfaces in both humans and animals[8]. *N. gonorrhoeae* is genomically, morphologically, and phenotypically closely related to the other pathogenic *Neisseria* species, *Neisseria meningitidis,* which is typically carried as a commensal in the (naso)pharynx of 10-15% of the general population but occasionally causes fatal septicaemia and/or meningitis[6,8-10]. *N. gonorrhoeae* is also related to several other commensal *Neisseria* species that reside particularly in the pharynx. Despite containing many of the pathogenicity and virulence factors of *N. gonorrhoeae* and *N. meningitidis*, the commensal *Neisseria* species, from which these two pathogenic *Neisseria* species have evolved, do not normally cause pathology[9], as they are unable to induce substantial polymorphonuclear leukocyte (PMNL)-based inflammation and lack several additional factors and mechanisms of interacting with host molecules, cells and tissues[11]. The pathogenesis and pathophysiology of *N. gonorrhoeae* have been studied for decades; however, detailed knowledge regarding many fundamental properties remains lacking.

The majority of men with gonococcal urethritis are symptomatic, but substantially fewer women with urogenital gonorrhoea are symptomatic and, when present, symptoms are non-specific. Nevertheless, signs of infection can be identified in most women with urogenital gonorrhoea. Rectal and pharyngeal gonorrhoea, which is mostly asymptomatic, are most frequently diagnosed in men-who-have-sex-with-men (MSM), but are not rare in women either. Disseminated gonococcal infections (DGI) are rare but can occur in both adults and neonates[6,12,13]. If infections are not detected and/or adequately treated, ascending infections, such as epididymitis and salpingitis, can result in a variety of serious complications and sequelae particularly in women, who bear the major burden of disease; these complications and sequelae include pelvic inflammatory disease (PID), chronic pelvic pain, ectopic pregnancy, and infertility. Gonorrhoea also facilitates the transmission and acquisition of other STIs including HIV infection. Gonococcal infections can lead to complications during pregnancy and infected women can also transmit infections to children during birth causing ophthalmia neonatorum, which was a leading cause of blindness in the pre-antimicrobial era. Conjunctivitis in adults is also observed sporadically. Consequently, gonorrhoea causes substantial morbidity and socioeconomic consequences globally[12,14,15].

In the absence of a gonococcal vaccine, management and control rely on effective, affordable, and accessible antimicrobial treatment, supported by adequate prevention, diagnostic testing or screening, notification and management of sex partners of infected individuals, and epidemiological surveillance. However, *N. gonorrhoeae* has developed or acquired antimicrobial resistance (AMR) to all antimicrobials earlier recommended as first-line or second-line empirical treatment of gonorrhoea, for example, sulphonamides, penicillins, tetracyclines, fluoroquinolones, and early-generation macrolides such as erythromycin. This extensive resistance has been accomplished by an accumulation of AMR determinants, most of which do not seem to substantially reduce the biological fitness of the bacterium (Figure 1)[16-21]. This AMR is of serious public health concern as the pathogen has become highly resistant to all previously recommended antimicrobials, and resistance to the currently recommended extended-spectrum cephalosporin (ESC) ceftriaxone and macrolide azithromycin has also emerged. On the basis of the high prevalence of gonorrhoea globally, high level of antimicrobial use and/or misuse, suboptimal diagnosis, limited control and surveillance of AMR, suboptimal or slow update of management guidelines, and the extraordinary ability of *N. gonorrhoeae* to acquire or develop —

and retain — AMR, it is likely that the global impact of gonorrhoea, including its severe complications and sequelae, will increase, and further *N. gonorrhoeae* AMR will evolve in the future. Consequently, improved global actions and research efforts to retain gonorrhoea as a readily treatable infection are essential.

This Primer focuses on the epidemiology, aetiological agent, pathogenic mechanisms/pathophysiology, diagnosis, screening, prevention, and management of gonorrhoea. We also discuss global actions and research efforts imperative for future management and control of gonorrhoea.

[H1] Epidemiology

In 2016, the WHO estimated that there were 86.9 (95% uncertainty interval: 58.6–123.4) million incident global cases of gonorrhoea (global prevalence: 0.9%) among adults of 15–49 years of age (Figure 2)[22]. The epidemiological diversity of gonorrhoea manifests itself in the variability in the geographical distribution and the prevalence among certain populations; determinants of such variability include sexuality and sexual orientation; socioeconomic, demographic, geographical and cultural ramifications (including stigma and taboos); and access to and quality of sex education, prevention, testing and diagnostics, as well as political commitment in the provision of health services[23-25].

[H2] Epidemiological determinants

When individual countries, especially in industrialised settings, embarked on prevention and care of STIs on the basis of the established determinants of STIs, declines in rates of gonococcal infections were observed during the late 1980s. However, this decline was short-lived, as increases in gonococcal infections rates have been reported since the late 1990s. Observations have identified a number of factors, both established and new, as important to explain the high rates of STIs, including gonococcal infections; these factors include ethnic background; sexuality and sexual preferences; sexual mixing patterns, such as assortative mixing by race and/or ethnicity (that is, the tendency to connect with individuals of the same race and/or ethnicity) and disassortative mixing by risk group (that is, the tendency to connect with individuals with a different risk level); gender and disparities in economic status and access to services, as well as the intrinsic characteristics of the pathogen[24,26-30].

Other reasons for the recent increase in gonorrhoea incidence in many high-resourced settings include changes in sexual behaviour in the era of antiretroviral treatment (ART) for HIV infection (that is, because of the availability of ART and the perception that HIV infection is no longer life-threatening in the short term, people are less cautious and have sex with new and casual partners without condoms), increased electronic connectivity (for example, the use of dating apps for meeting sex partners), increased number of casual unknown partners, larger sexual networks, increased travel, and variable access to services[30,31]. Another factor to be taken into consideration is the increasing use of drugs in sexual networks, particularly common among MSM and female sex workers. Finally, certain key populations are at higher risk for and disproportionately affected by STIs, including gonorrhoea; such populations include MSM, migrants, young people and sex workers.

[H2] Incidence and prevalence

The aforementioned factors, mostly in combination, probably substantially contribute to the varying increases in gonorrhoea case rates in the past 5–10 years, even in countries with more comprehensive health systems. For example, in the USA and in the European Union/European Economic Area (EU/EEA), both socioeconomic status and ethnic background have been observed to highly correlate with gonococcal infection rates. In the USA in 2017, the rate of reported cases of gonorrhoea was ~8 times higher among black populations than among white counterparts. Higher rates were also noted among American Indians and Alaska Natives, Native Hawaiians and individuals with Hispanic heritage, whereas the rate among individuals with Asian heritage was half the rate among white individuals[30,31]. In the USA, the number of gonorrhoea cases increased by 67% from 2013 (n=333,510) to 2017 (n=556,413)[32]. The proportion of gonococcal isolates cultured from MSM increased from 3.9% in 1989 to a high of 38.5% in 2017, reflecting epidemiological changes and possibly changes in the healthcare-seeking behaviour of men with gonorrhoea as well as improved reporting of sexual orientation in the USA[30,31].

In the EU/EEA, the number of reported gonorrhoea cases has increased by >200% since 2008, from 29434 cases in 2008 (with an incidence of 7.85 per 100,000 population) to 89239 cases in 2017, with the highest numbers of cases in the UK, France, The Netherlands, and Spain[33]. Of note, higher prevalence in these countries might be in part accounted for by the availability of comprehensive sexual health systems, frequent testing and/or surveillance. The highest incidence of gonorrhoea in EU/EEA is in young adults (15–24 years of age)[33]. MSM accounted for about 25-30% of all the cases in the EU/EEA during recent years — 30% of the reported gonorrhoea cases (57% of the cases reporting sexual orientation) in Europe in 2017 (Ref[33]); however, over the past decade substantial increases also occurred among heterosexual men, men with no sexual orientation reported, and women. In the UK, MSM experienced substantial increases in reported STIs in 2017. Of the 50,032 new non-viral STI diagnoses in MSM in 2017, 43% were gonococcal infections, and, between 2016 and 2017, gonococcal infection diagnoses increased by 21%[34].

The geographical setting in which people live also seems to have a role in the prevalence of gonococcal infection, probably reflecting differences in the access to information regarding STIs; availability, accessibility and quality of health care services; and social factors such as the effect of stigma on health seeking behaviors. Observations showed that the prevalence of gonorrhoea in women of 15–24 years of age in clinical or community settings in South Africa was ~4.6%, whereas in southern Africa and eastern Africa the prevalence was 1.7%. Furthermore, in the same study, the prevalence in a high-risk population in eastern Africa, mostly sex workers, was 8.2%[35].

In low-income settings, mainly syndromic management of STIs is performed, and there are no comprehensive aetiology-based surveillance systems that would enable an accurate assessment nation-wide of increases or decreases in gonorrhoea prevalence in the general population or in subpopulations. However, even in many high-income settings, for example in Europe, the surveillance data should be interpreted with caution as the surveillance systems, testing, methodologies, and quality assurance are not standardized across countries and remain weak in several settings[33,36]. Finally, whole genome sequencing (WGS) will revolutionize our understanding of the epidemiology of gonorrhoea and the geographical and temporal spread of AMR and antimicrobial susceptible *N. gonorrhoeae* strains in different populations and subpopulations, including at-risk groups (see the Outlook section).

[H3] Gonorrhoea in MSM on pre-exposure prophylaxis

Another topical area of interest is the observation of rapid increases in the incidence of gonorrhoea, and other STIs, in high-resourced settings among MSM taking pre-exposure prophylaxis (PrEP) for prevention of HIV infection. Some published data reported that MSM using PrEP can be ~25 times more likely to acquire a gonococcal infection than MSM not using PrEP[37]. A multisite open-label study of just under 3,000 gay and bisexual men using PrEP, conducted in Australia between 2016 and 2018, showed significant increase in incidence of STIs (including gonorrhoea, *Chlamydia trachomatis* infection and syphilis), during a follow-up period of 1.1 years. Younger age, greater number of sex partners and group sex participation were associated with greater risk for an STI, whereas inconsistent or no condom use with casual partners was not[38]. A systematic review commissioned by the WHO in 2018-19 identified 88 STI studies, primarily in MSM in high-income countries, which found that STIs prevalence was high in people prior to starting PrEP, and STIs incidence varied by setting and population included in the review. However, pooled STIs incidence generally remained high during follow-up when taking PrEP[39,40]. It should be noted, however, that persons on PrEP are monitored more closely and tested more frequently for STIs than non-PrEP users. When both populations were controlled for frequent monitoring, as in the PROUD study, no statistically significant differences in STIs rates were found between men taking PrEP and the control group[41]. Thus, it would seem that the reduced risk for and fear of HIV infection have led some PrEP users especially young MSM, to reduce condom use and/or increase other risky sexual behaviours, and, therefore, to place themselves at increased exposure to other STIs, including gonorrhoea. However, given the conflicting conclusions from different population studies on this point, more observations and studies are needed to identify the factors behind these contradictory conclusions, as well as to detail the risk factors and elements that may be responsible for the findings of increased STI risk in some populations and to better understand the ideal monitoring and screening intervals of individuals taking PrEP.

[H1] Mechanisms/pathophysiology

[H2] The bacterium *Neisseria gonorrhoeae*

[H3] Growth and metabolism

N. gonorrhoeae is a fastidious organism, sensitive to many environmental factors such as oxygen, non-physiological temperatures, desiccation, and presence of toxic substances (such as many fatty acids), among others[42]; thus, the bacterium does not survive for long outside the human host, and is difficult to culture (box 1). Many strains have incomplete biosynthetic capabilities for amino acids, presumably because amino acids and other important nutrients are readily obtained from the human host. Iron (which is essential for bacterial growth) is acquired from the host by binding iron-containing host proteins like transferrin, lactoferrin, and haemoglobin at the bacterial surface and stripping these molecules of iron that is then delivered to the bacterial cytoplasm[43]. Owing to the broad range of oxygen levels within different niches of the male and female urogenital tracts, it is possible that *N. gonorrhoeae* encounters aerobic, microaerobic, and anaerobic conditions within the host, and the bacteria are able to grow in all these conditions[44].

[H3] Genetics

Using WGS, it has been shown that the modern gonococcal population is not as old as previously considered and has been shaped by antimicrobial treatment of STIs as well as other infections, leading to the emergence of two major genomic lineages, one multidrug-resistant and one multidrug-susceptible, with different evolutionary strategies[45] *N. gonorrhoeae* has a single circular chromosome between ~2.1 and 2.3 megabase pairs (~2200-2500 protein coding sequences), which exists as diploid, homozygous, chromosomes[46,47]. In addition, *N. gonorrhoeae* can acquire additional DNA via horizontal genetic transfer (HGT), the non-inherited external acquisition of new genetic material from another bacterium. HGT occurs mainly by Type IV pilus-mediated DNA transformation (uptake of DNA from the environment and subsequent incorporation into the genome). *N. gonorrhoeae* is naturally competent for transformation during its entire life cycle, but transformation only occurs at high frequency between cells of *N. gonorrhoeae* and other *Neisseria* species. Approximately 80% of isolates carry a chromosomal insertion called the gonococcal genetic island, which has genes similar to those carried on conjugal plasmid, that is, genes involved in conjugation (the DNA transfer between bacteria by cell-to-cell contact). However, in *N. gonorrhoeae* these conjugation gene products act to secrete chromosomal DNA into the medium that is then available for DNA transformation. Pilus-mediated DNA transformation provides efficient transport of DNA into the bacterial cell and the DNA uptake sequences highly represented in *Neisseria* genomes (~1900-2000 copies per genome)[48,49]. This efficient transformation is one reason why AMR determinants efficiently spread from cell to cell. Notably, this ability of *N. gonorrhoeae* to transfer DNA between strains makes clonal analysis difficult since alleles are not stably linked and led to the creation of the Multi Locus Sequence Typing (MLST) system to characterize bacterial lineages by the DNA sequence type of several defined and more conserved housekeeping genes[50]. MLST systems are now available for many different bacterial species[51]. Furthermore, this re-assortment of alleles suggests that mixed strain gonorrhoea infections are common[52,53], although widely unrecognized, as most clinical laboratories analyze and save single colonies when culturing isolates, probably underestimating the incidence of mixed infections. Ideally, multiple colonies should be tested.

Nearly all gonococcal strains contain a cryptic plasmid (with no defined functions), and many contain a plasmid encoding a penicillinase (mostly TEM-1 or TEM-135 β-lactamase), which results in high-level penicillin resistance, and conjugative plasmids, which sometimes carry *tetM* causing high-level tetracycline resistance, although these plasmids are not as prevalent as reported for many other bacterial species[16,54,]. Several penicillinase-encoding plasmids of different size have been described in *N. gonorrhoeae* and named according to their epidemiological origin, such as the widely spread and most common African, Asian, and Rio/Toronto plasmids. Different conjugative gonococcal plasmids carrying *tetM* have also been described, the most common being the American *tetM* plasmid and the Dutch *tetM* plasmid[16,54]. In addition, several double-stranded and single-stranded bacteriophage gene islands have been annotated within the *N. gonorrhoeae* genome, but no isolated bacteriophage that can infect and lyse the bacteria has been found[55].

[H3] Colonisation determinants

N. gonorrhoeae shares many colonisation determinants with other human-restricted *Neisseria* species that rarely cause infection. The factors required to establish a host niche include the Type IV pilus, the opacity protein family (Opa proteins), the porin PorB, efflux pumps, and metal transport systems (Figure 3). *N. gonorrhoeae* probably has to compete with the resident microbiota for colonization, but little is known about how different resident commensal organisms may limit or cooperate with *N. gonorrhoeae* during colonization.

Gonococcal pili are required for efficient mucosal colonization (typically of non-ciliated columnar epithelium) and carry out many functions including: initial adherence to host cells and tissues, self-adherence and adherence to other *N. gonorrhoeae* cells, a means to crawl along mucosal surfaces called twitching motility, protection from PMNL killing mechanisms[56], and HGT by DNA transformation[57]. Clinical isolates of *N. gonorrhoeae* are always piliated, but quickly lose pilus expression in laboratory culture through a variety of mechanisms, showing that pilus expression is under strong selective pressure during infection.

The Opa proteins mainly act as adhesins that bind to a variety of receptors found on many different cells and tissues[58] and mediate more intimate attachment and initiation of microcolony formation. Most Opa proteins bind to one or more human carcinoembryonic antigen-related cell adhesion molecules (CEACAMs), a family of surface-exposed proteins. Opa proteins only bind to human forms of these proteins, and a few Opa proteins also bind to heparan sulfate proteoglycans. While some Opa-CEACAM interactions lead to cell signaling events, such as induction of the oxidative burst from PMNLs, most Opa interactions seem to be important for adherence to cells and tissues[59].

All Gram-negative bacterial porins (transmembrane channel proteins) act to allow small molecules access to the periplasm. The *N. gonorrhoeae* porin (PorB) is one of the most abundant proteins in the outer membrane: it increases attachment, is then translocated to the host cell mitochondria, and impairs the ability of phagocytes to kill the bacteria. Other important properties include resisting the action of complement factors, modulating apoptosis, invasion of host cells, and involvement in AMR[60-63].

N. gonorrhoeae expresses up to five efflux pump systems: MtrC–MtrD–MtrE, MacA–MacB–MtrE, NorM, FarA–FarB–MtrE and MtrF[64-66]. These export pumps have varying narrow or extensive substrate specificity and have many roles in pathogenesis, including removing toxic molecules encountered during infection, like fatty acids and cationic peptides, and removing antimicrobials from the cell, that is, acting as AMR determinants. Finally, there are three iron acquisition systems in the envelope of *N. gonorrhoeae*, and each can strip iron from a human protein that is designed to sequester iron from pathogenic organisms. There is an acquisition system for transferrin (TbpA-TbpB), one for lactoferrin (LbpA-LbpB), and one for haeme (which can be found, for example, in haemoglobin) (HpuA-HpuB)[43].

[H2] Infection dynamics

All bacteria that live in or on people need to colonize and grow, whether they are commensal organisms that rarely cause harm or frank pathogens. The pathogenesis field defines colonisation and growth determinants as virulence determinants even though they are often found also within organisms that do not cause overt pathology. However, for a pathogenic organism to do damage, it usually needs to colonize specific anatomical sites and grow (except when pathogenesis occurs through production of a toxin away from the site of infection).

[H3] Transmission

N. gonorrhoeae infects the mucosal epithelium of the male and female urogenital tracts, the rectum, pharynx, or conjunctiva[12]. *N. gonorrhoeae* is mainly transmitted through unprotected vaginal, anal or oral intercourse. During vaginal sex, transmission rates from men to women are higher than from women to men[67]. Ejaculate from infected men contains millions of bacteria, effectively injecting the organism into the receiving anatomical site. How the organism is effectively transmitted from vaginal, rectal, or oral/pharyngeal locations to the male urethra is not completely understood. Of note, *N. gonorrhoeae* infection amplifies the risk for acquisition and transmission of HIV and several other STIs[68,69]: all the underlying mechanisms are not completely understood, but probably involve factors such as inflammation, destruction of the mucosa, and discharges. Furthermore, women with *N. gonorrhoeae* infection can effectively transmit the infection to their children during birth (intra-partum), but not during pregnancy; the neonate's conjunctiva is highly exposed during transit of the birth canal, and *N. gonorrhoeae* infection of the conjunctiva results in ophthalmia neonatorum.

Host defenses against infection act at many levels. *N. gonorrhoeae* has no ability to persist on or penetrate the skin, and requires a mucous membrane for colonisation. Many barriers in mammalian cells limit transit of organisms into the body, including the ciliary action of some epithelia. Peptidoglycan fragments and lipooligosaccharides (LOS) released by *N. gonorrhoeae* can disrupt the ciliary action of the epithelium and may promote colonisation[70,71]. Once colonisation is established, innate and adaptive immune responses act to block or limit the growth of an organism. However, as a host-restricted organism that has co-evolved with its human host, *N. gonorrhoeae* has intricate mechanisms to limit the action of these host defense systems.

[H3] Innate immune systems

Resident tissue macrophages are one of the first cells that *N. gonorrhoeae* encounters during infection (Figure 4)[72]. Whether macrophages have a role in limiting *N. gonorrhoeae* infection is not clear, but macrophages, dendritic cells and epithelial cells may all be responsible for producing the chemokines and cytokines induced during infection. Some of these host effectors are responsible for inducing the massive PMNL response that manifests as the purulent exudate characteristic of symptomatic urethral gonorrhoea. *N. gonorrhoeae* can survive the various antimicrobial functions of PMNLs including phagocytosis; the release of reactive oxygen species, cationic peptides and antimicrobial enzymes; metal sequestration; and PMNL extracellular traps[73]. *N. gonorrhoeae* can also modulate the apoptosis of epithelial cells, macrophages, T cells, and PMNLs, but since both the inhibition and enhancement of apoptosis has been reported, the relevance of apoptosis modulation to infection remains controversial[74,75]. In addition, the role of PMNLs during *N. gonorrhoeae* infection also remains controversial. PMNLs probably influence infection by killing some of the bacteria but allowing the spread of others[73].

The classical and alternative complement pathways act to kill many organisms, and *N. gonorrhoeae* has evolved ways to avoid both pathways during uncomplicated infections[76]. Indicative of its extreme host restriction and evolution, *N. gonorrhoeae* remains sensitive to animal complement system components[61]. There are several mechanisms *N. gonorrhoeae* uses to limit complement-mediated killing by blocking deposition or activity of several complement factors (Figure 4)[61]. People with complement deficiencies are at increased risk of DGI, showing that the complement system helps to limit gonococcal survival in the blood stream[77]. Increased incidence of DGI and other disseminated *Neisseria* spp. infections was observed when patients were treated with eculizumab, a complement inhibitor, but this study did not report altered rates of uncomplicated gonorrhoea[78]. It is not fully known whether complement effectively functions at mucosal sites of colonization.

[H3] Adaptive immunity

As an organism that has co-evolved with its sole host for centuries, and possibly throughout all recorded time, *N. gonorrhoeae*'s colonisation determinants are exquisitely adapted to life within humans. By contrast, the human adapted immune system has variable components (B cells and T cells) that can change to limit infection. *N. gonorrhoeae* is generally thought to be immunosuppressive[79], although there are suggestions that any immunosuppression is incomplete. Many studies show that anti-gonococcal antibodies are found in people with active or previous infection, demonstrating a humoral immune response[80]. In addition, the existence of three, independent, antigenically-variable surface antigens (type IV pilus, Opa proteins, and LOS) also provides evidence that there are potentially protective responses directed against these antigens that necessitates the complex variations[81]. These antigens can all vary during infection and colonization, for example, the surface exposed antigenic epitopes of pili will vary and pilus expression can be lost, the number and type of expressed Opa proteins will vary (Figure 3), and the type of sugars on the LOS molecule can change. While some of this surface variation alters some functional properties of *N. gonorrhoeae*, the most important function of antigenic variation is immune avoidance, which enables reinfection presumably even with the same gonococcal strain, as protective immunity to *N. gonorrhoeae* capable to prevent subsequent infections has never been recorded. Extensive surface molecule variation by *N. gonorrhoeae* also prevents these molecules from being considered viable vaccine candidates. A more detailed examination of immune suppression and responses during human infection is needed.

[H3] Host damage

N. gonorrhoeae is not a very disruptive pathogen, as it is well-adapted to its human host and rarely lethal. It does not produce any exotoxins that can destroy host cells, but does secrete peptidoglycan fragments, outer membrane vesicles (OMVs) and LOS that are toxic to mammalian cells and can specifically inhibit the ciliated cells on fallopian tube tissues[70,71]. Moreover, when PMNLs are recruited to sites of infection, PMNL antimicrobial products are released that can damage the tissue. All of these factors contribute to the damage and scarring of the fallopian tube tissue that is characteristic of PID. These factors can also cause damage at other sites of infection, particularly during DGI, in which, in addition to fever, also dermatitis, infectious arthritis and less frequently septicaemia, endocarditis and meningitis can occur.

[H1] Diagnosis, screening and prevention

[H2] Clinical presentation and diagnosis

The incubation period for urogenital gonorrhoea ranges from ~2 to 8 days[82]. The clinical manifestations of gonorrhoea are variable and differ markedly in men and women[12]. At least 90% of men with gonococcal urethritis are symptomatic, presenting with obvious urethral discharge and dysuria, a fact that permits the application of syndromic diagnosis (based on a set of symptoms and signs that are characteristic of a clinical manifestation) in many settings as both a time-saving and cost-saving measure. For men with symptomatic urethritis, Gram stain may be used to support symptom evaluation. By contrast, laboratory-based diagnostic tests have a more important role for gonococcal detection in asymptomatic men, women and in patients of all genders for extragenital (rectal and pharyngeal) infections, which are mostly asymptomatic or present with non-specific symptoms. Although ~40% of women with gonococcal cervicitis may report abnormal vaginal discharge, this symptom is unreliable for syndromic diagnosis of gonorrhoea in women, as many other equally or more common genitourinary infections in women (for example, bacterial vaginosis, trichomoniasis and vaginal candidiasis) may cause the same symptoms.

Microbiological diagnosis of gonorrhoea can be challenging, as many regions do not have laboratory-based diagnostic capability and rely on syndromic management algorithms to guide empiric antimicrobial treatments[14]. Microbiological diagnosis is performed by detection of Gram-negative diplococci in stained smears using microscopy, culture of *N. gonorrhoeae*, and/or nucleic acid amplification tests (NAATs) detecting *N. gonorrhoeae* DNA or RNA.

[H2] Traditional diagnostic methods

[H3] Microscopy

In resource-limited settings, light microscopy of especially Gram-stained samples is often the only method available to diagnose infection with *N. gonorrhoeae* presumptively (Table 1). The sensitivity and specificity of the Gram stain, which tests for the presence of characteristic Gram-negative diplococci within PMNL, can vary substantially between studies and depends upon the specimen: the highest sensitivity and specificity were reported with urethral swabs samples from symptomatic males (89% to >98% and >95%, respectively)[6,13,83-85], whereas the sensitivity was as low as 40-50% in urethral specimens from asymptomatic males, and in endocervical or urethral specimens from women[13,83,84]. This difference can probably be explained by a reduced bacterial load particularly in these urethral samples and additionally the presence of many other bacterial species in the endocervical samples. Gram stain is not suitable for the diagnosis of *N. gonorrhoeae* from pharyngeal specimens (because other *Neisseria* species with similar morphology are prevalent in the oral and nasopharyngeal cavity) or rectal specimens (which have a sensitivity ≤40%)[82-84]. A methylene blue staining method is an alternative to the Gram stain and similar high sensitivity and specificity were reported for diagnosing gonococcal urethritis in men[86].

[H3] Culture

Prior to the introduction of NAATs, culture (Table 1) of the organism was the gold standard and this remains the only diagnostic method available in some settings, as it is low-cost. Culture also remains recommended for test of cure (TOC) for treatment failure; in cases of sexual abuse; and to evaluate PID[13,85,87]. Furthermore, complete AMR testing can only be accomplished if *N. gonorrhoeae* is cultured[83,85,87,88]. Culture performance is dependent upon factors such as anatomical site of the cultured sample, method of specimen collection, media and conditions used to transport the sample to the diagnostic centre[83,87,89], non-selective and/or selective culture media[84,85,89,90], conditions of incubation[82,85], and species confirmatory tests. Cultures obtained too soon after exposure (under 48 hours) may give false negative results[13], and a repeated culture sample some weeks later is sometimes considered. Culture of urogenital specimens usually has a sensitivity ranging from 72-95%, but can have a sensitivity of 95-100% in settings with extensive experience in appropriate specimen handling and culture[83,84]. However, the sensitivity of culturing pharyngeal and rectal specimens is much lower.

Presumptive identification of cultured *N. gonorrhoeae* isolates is frequently accomplished by typical colony appearance on selective media, Gram-stained microscopy, and the oxidase test, which detects the presence of cytochrome oxidase[82,84,85]. For definitive *N. gonorrhoeae* identification, immunological tests frequently targeting PorB[85,91-93]; sugar utilization tests or other biochemical tests[6,85,91,94]; NAATs, or Mass Spectrometry (that is, matrix-associated laser desorption ionization time of flight (MALDI-TOF))[6,95-97] are frequently performed. These tests differentiate *N. gonorrhoeae* from species such as *N. meningitidis*, *N. lactamica*, *N. cinerea, N. subflava*, or other genera that occasionally may grow on even the selective culture media and may be present particularly in the pharynx but also at other sites[85]. Finally, DNA extraction from cultured isolates is also currently the best method to obtain DNA for genomic analysis, as clinical specimens often either do not contain sufficient concentrations of DNA, or contain too much DNA from other bacterial species or human cells. Furthermore, methods for genomic DNA purification from clinical specimens have not been sufficiently developed or standardized[98].

[H2] NAATs

NAATs are currently recommended for gonorrhoea diagnosis in most high-income countries[13,82,87,99]. NAATs are now the preferred diagnostic test because specimen collection is non-invasive (urine or self-collected swabs); viable organisms are not required for detection, permitting less stringent transportation and storage methods[85,100]; most have superior sensitivity with maintained high specificity (which vary between NAATs and anatomical site tested) compared with culture; they produce more rapid results (many later generation NAAT platforms allow for high throughput and automation); and many can simultaneously detect other STI-associated pathogens (particularly *C. trachomatis*)[13,87,101,85]. Initially, a number of in-house, PCR-based NAATs were used locally and continue to be used as confirmatory tests or for diagnosis in resource-limited settings[93,102-104]. In-house NAATs generally target conserved regions of genes such as the the *porA* pseudogene, *opa* genes, *gyrA* (encoding DNA gyrase subunit A), *cppB* (encoding cryptic plasmid protein B) and the methyltransferase genes of *N. gonorrhoeae*[102]. Few reports compare the performance of such in-house NAATs with culture and especially commercially available NAATs[102]. In high-income countries, in-house NAATs have largely been replaced with commercial NAATs that have been comprehensively validated and received regulatory approval from the US Food and Drug Administration (FDA) (Table 2)[13,87,101].

In 2019, the first two NAATs (Aptima Combo 2 assay and Xpert CT/NG) for gonococcal detection received FDA approval also for extragenital specimens such as rectal and pharyngeal infection[105], and licensing for additional NAATs is in progress. Several studies indicate that many additional NAATs are more sensitive, with maintained high specificity, than culture for diagnosing *N. gonorrhoeae* from pharyngeal and rectal specimens (Table 2); however, such tests should be used only after rigorous local performance evaluations[82,87,106], and additionally a confirmatory NAAT with a different target should be used for such specimens[82,87,100], as other *Neisseria* species, which can be frequently present especially in pharynx, could be misidentified as *N. gonorrhoeae*[87,100]. Thus, when using NAATs to detect *N. gonorrhoeae*, it is important to choose the test or the testing strategy so that the positive predictive value (PPV, which is calculated based on the sensitivity and specificity of the test and on the local prevalence of the pathogen, and the last two parameters substantially affect the PPV) is >90%[82,85].

The introduction of NAATs for *N. gonorrhoeae* has substantially reduced the number of cultured patient samples. FDA-approved NAATs are more expensive than culture-based methods, and mostly utilized in high-income countries[13,82,87,99]. Pooling specimens (that is, combining up to 5–10 specimens and then retesting them separately if the pool is positive to ascertain which specimen(s) was positive) may reduce cost, especially in settings with high-volume testing and with low positivity rate. However, strict evaluation of the performance characteristics of the NAAT in the local population is crucial before implementing any pooling strategy. Time to results, hands-on time, maintenance and consumption of reagents and consumables for automated platforms vary greatly between platforms, and these parameters influence the choice of platform[107,108]. A major disadvantage of commercial NAATs is the inability to perform AMR testing on gonococcal specimens[14,85,102,109]. In many regions, >80% of gonorrhoea cases are diagnosed by NAATs and, therefore, crucial information regarding AMR and gonococcal strain biology is lost. There are no recommended molecular tests for the prediction of antimicrobial susceptibility or resistance[102,110,111]; however, a PCR-based test that also detects ciprofloxacin susceptibility status has received CE-IVD Mark (Table 3) and several NAATs in the pipeline are also being developed to detect both *N. gonorrhoeae* and its ciprofloxacin susceptibility status[101]. This type of test could be important particularly in regions in which ciprofloxacin susceptible strains are still spreading, and, therefore, ciprofloxacin could be used for treatment as a lower cost oral alternative to ceftriaxone plus azithromycin, that is to spare the use of these antimicrobials and accordingly decrease the selective pressure for resistance. This concept has been tested clinically with success[101,112,113]. Notably, both the British Association for Sexual Health and HIV (BASHH) gonorrhoea guideline for the United Kingdom and the European gonorrhoea guideline for the WHO European Region recommend use of ciprofloxacin for treatment of anogenital and pharyngeal gonorrhoea if the gonococcal strain causing the infection is proven ciprofloxacin susceptible using genetic or phenotypic resistance testing[82,114].

[H2] Point-of-care tests (POCTs)

Development of appropriate rapid point-of-care tests (POCTs) is a high priority for the diagnosis of gonorrhoea[14,85,101,115] (Table 3). POCTs could provide a definitive, rapid diagnosis to guide specific treatment in situations where this is not currently possible, such as in settings in which only syndromic management is available or in cases where patients may not return for treatment and for screening asymptomatic patients[116-118]. Ideally, POCTs should meet the 'ASSURED' criteria, that is, be affordable, sensitive, specific, user-friendly, robust and rapid, and equipment free (or requiring minimal equipment powered by solar or battery sources)[117,119,120], but all diagnostic tests that provide rapid test results and correct treatment during a single clinical visit could be defined as POCTs[117,121,122]. The Gram stain is an oft-used POCT; its limitations have been described above[122,123]. Other POCTs developed for *N. gonorrhoeae* include lateral flow immunochromatographic (ICT) and optical immunoassay (OIA) tests based on antigen detection, as well as a near-POCT NAAT — the Xpert CT/NG assay[101,120,122,123]. Recent reviews of the performance of several POCTs have shown that ICT-based and OIA-based POCTs had highly suboptimal sensitivities, some as low as 12.5%, and specificities ranging from 89% to >97%[120,123] and, therefore, are not recommended. However, mathematical modelling has shown that the sensitivity required for POCTs to be effective may be lower in settings where there is a high risk for transmission because treatment is delayed pending testing results or patients do not return for treatment[124]. The Xpert CT/NG assay has been successfully implemented as a near-POCT in areas such as Papua New Guinea, South Africa and remote regions of Australia[6,101,115,125,126]. However, this test is expensive, needs substantial electricity, and results take ~90 minutes.

[H2] Screening and prevention

Screening general populations for gonococcal infections is not indicated. However, screening or opportunistic testing can be considered for individuals at risk of gonococcal infection. These populations include: sexually active youth, sexual contacts of persons having a suspected gonococcal infection, MSM, persons with new or multiple sexual partners, persons with HIV infection or a history of STIs, sex workers and their sexual partners, and women (≤35 years of age) and men (≤30 years of age) at initial admission to a correctional facility[6,13,83,127,128]. The US CDC guidelines recommend annual screening for gonorrhoea of all sexually active females of <25 years of age and older women at increased risk of infection, and screening should also be offered to young MSM[127,128]. More recently, in the US, owing to observed high rates of incident infections, screening for gonorrhoea and other bacterial STIs (*C. trachomatis* infections and syphilis) has been recommended at 3-6 month intervals for persons receiving HIV PrEP[129]. In other high-income settings, there are no screening recommendations for general population owing to the low cost-effectiveness and low population prevalence of gonorrhoea, which results in low positive predictive values of the testing and increased probability of false positive results, which could cause considerable harm for patients and their partners. No aetiologically-based screening is performed in any low-income settings.

Main prevention efforts include education regarding symptomatic and asymptomatic gonorrhoea and other STIs; promotion of safe sexual behaviours (for example, increase condom use through condom-promotion education and campaigns); behaviour change communication programmes (for example, promoting fewer unknown, casual and unprotected sexual contacts and early health seeking behaviour); improved sexual partner notification and treatment; and expansion of targeted interventions, including screening in some settings for vulnerable populations (sex workers, MSM, adolescents and patients with STIs and their sexual partners)[130].

[H3] Vaccines

Given the threat of untreatable gonorrhoea due to the spread of AMR and the high burden of gonorrhoea worldwide, the need for a gonococcal vaccine has become increasingly urgent[131-133]. Prior to the 1990s, four vaccine candidates progressed to clinical trials: a whole cell vaccine, a partially autolyzed vaccine, a pilus-based vaccine, and a PorB-based vaccine[133-135]; none provided much protection from infection. Gonococcal vaccine development is complicated by the biology of the gonococcus. Limitations include the scarce adaptive immune responses to gonococcal infections, lack of known correlates of protection, antigenic variability of the potential vaccine candidate antigens, production of blocking antibodies (which upon binding their target prevent the binding of other antibodies — for example, bactericidal antibodies — to the same target or other targets in close proximity) to conserved antigens, and lack of robust, small laboratory animals for testing vaccines[132,134].

However, recently, it has been noted, in several countries, that there was a decline in the number of gonorrhoea cases following the use of meningococcal group B OMV vaccines against *N. meningitidis*[136]. One of these vaccines, with the trade name MeNZB, was associated with reduced rates of gonorrhoea diagnosis and of hospitalization from gonorrhoea[136], and it seems to provide proof-of-principle to inform the development of gonococcal vaccines[137,138]. Research to elucidate the specific or nonspecific antigens and mechanisms involved in the MeNZB-mediated protection against gonorrhoea is crucial. MeNZB is no longer available, however, the licensed, four-component meningococcal group B vaccine 4CMenB (trade name BEXSERO;

GlaxoSmithKline) includes the same OMV as MeNZB and three recombinant meningococcal antigens (Neisserial heparin-binding antigen (NHBA), Factor H-binding protein (fHbp), and Neisseria adhesin A (NadA)), which are also relatively conserved compared with their gonococcal homologues[139]. Accordingly, high coverage of the 4CMenB in the population may also decrease gonorrhoea prevalence. Recently research has exploited OMVs from *N. meningitidis* expressing factor H-binding protein and found that serum bactericidal antibodies against the gonococcus were produced in mice, although sera from humans immunized with 4CMenB were not bactericidal for *N. gonorrhoeae*[140]. These findings together with the immunobiology research (including on *N. gonorrhoeae* immune suppressive responses and how they can be overcome), antigen discovery and animal modelling are promising for vaccine development.

[H1] Management

[H2] Management principles

Gonorrhoea is a community-based infection and often there is limited follow-up after treatment. Prompt and effective treatment reduces complications and eliminates transmission of the infection[128]. Since there are no vaccines and host immunity cannot prevent reinfection, eradication of infections is solely reliant upon case finding and ideally microbiological diagnosis coupled with effective antimicrobial treatment[128]. Of note, because gonorrhoea also amplifies risk for acquisition and transmission of HIV, gonorrhoea control also contributes to global efforts to reduce HIV infections. The goal of gonorrhoea management is to quickly and accurately identify infected persons, enabling provision of timely treatment to prevent complications and transmission of infection to sexual partners and, for pregnant women, to children at the time of birth. Factors influencing management include considerations of the clinical manifestations, the disproportionate morbidity for women (PID, infertility, ectopic pregnancy, chronic pelvic pain), and stigma associated with STIs. As the infection is most common in resource-limited settings (even in high income nations, gonorrhoea is most common among marginalized populations who may have limited resources and/or limited access to health care), costs of both diagnosis and treatment may also influence the translation of management principles into practice.

Because gonorrhoea transmission most often is a consequence of sex with a person who is unaware of his/her infection, notification, testing and treatment of recent sexual partners is a crucial part of gonorrhoea management within communities[82,141]. Notification and referral of exposed sexual partners of persons with STIs (by health care providers, public health specialists or the partner himself/herself) has been recommended since at least the 1940s[142]. However, programmes promoting notification of sexual partners have often proved resource intensive and failed to successfully lead to treatment of many sexual partners, probably in part owing to stigma and embarrassment regarding having an STI. Thus, “expedited partner therapy” or “partner-delivered therapy” (that is, the partner(s) of a patient with gonorrhoea receive oral, single dose antimicrobials delivered by the patient, without have being examined or tested) for gonococcal and chlamydial infections has been increasingly practiced in USA with good results[143]. Currently, cefixime plus azithromycin is used for expedited partner therapy for heterosexual men and women[128]. However, this approach has raised concerns about the lack of clinical examination, lack of testing for additional STIs, lack of opportunities to trace ‘downstream’ sex partners, possible antimicrobial allergy or adverse events experienced by the partner(s) and AMR emergence.

[H2] Antimicrobial therapy

Syndromic management of urethral discharge in men can be relatively effective for gonorrhoea[116]. However, appropriate, local and aetiologically-based studies to regularly refine the syndromic management algorithm(s) are imperative, and nevertheless some infections (for example *C. trachomatis* and *Mycoplasma genitalium* infections) cannot be distinguished from gonorrhoea, resulting in overtreatment. Syndromic management of vaginal discharge both fails to detect and treat the substantial proportion of asymptomatic infections in women (who might continue to transmit the infection) and leads to vast overtreatment of symptomatic women who do not have gonorrhoea but *C. trachomatis*, *M. genitalium* or *Trichomonas vaginalis* infection or bacterial vaginosis[109,116].

Single-dose directly-observed systemic therapy (as topical therapy has not proved effective) that is provided in the care setting is preferred, to assure medications are delivered. Dual antimicrobial therapy (mainly parenteral ceftriaxone plus oral azithromycin) is currently recommended for empirical first-line therapy by the WHO global guidelines[109] and in most high-income countries, including European countries[82], USA[128], Canada[144] and Australia[145]; however, in some countries (for example, Japan[146] and, since 2019, the United Kingdom[114]) ceftriaxone high-dose (1 g) monotherapy is recommended[147-149]. In some international and national guidelines, cefixime plus azithromycin is recommended as an alternative regimen, but only if ceftriaxone is not available or the injection refused[82,128]. There is an ongoing debate among experts as to whether single or dual antimicrobial therapy should be the recommended therapy for uncomplicated gonorrhoea. The rationale for introducing dual therapy was to address the problem of *C. trachomatis* co-infection, which occurs in 10-40% of persons with urogenital gonorrhoea[150], as well as a hypothetical benefit of reducing the emergence and spread of AMR (particularly resistance to ceftriaxone) in *N. gonorrhoeae*. When possible, well tolerated oral therapy is preferred by both patients and clinicians[151]. Finally, persons with gonorrhoea are often co-infected with other pathogens, including *Chlamydia trachomatis*, *Trichomonas vaginalis*, *Treponema pallidum* and/or *M. genitalium* and, therefore, require treatment either with agents that are also effective against these pathogens or with co-therapy.

The continuing development of AMR by the gonococcus, coupled with a diminished pipeline for development of new antimicrobials have narrowed available therapies for gonorrhoea to a single agent that is sufficiently effective for first-line monotherapy, that is, parenteral ceftriaxone[16,152], which is frequently given together with azithromycin. If ceftriaxone is unavailable, or the patient has β-lactam antimicrobial allergy or is infected by a ceftriaxone-resistant gonococcal strain, therapy is challenging and highly variable, often utilizing ciprofloxacin monotherapy (if the gonococcal strain causing the infection has been proven susceptible by phenotypic or genetic resistance testing[82,114]), high dose (2 g) azithromycin monotherapy, spectinomycin (together with high dose azithromycin, particularly if pharyngeal gonorrhoea has not been excluded), or gentamicin (together with high dose azithromycin, particularly if pharyngeal gonorrhoea has not been excluded)[82,128]. However, each of these alternate therapies has limitations related to gonococcal resistance, antimicrobial availability and patient tolerance. Progressive decreases in susceptibility of *N. gonorrhoeae* to ceftriaxone, as well as to other antimicrobials, create a pressing need for continued monitoring of gonococcal AMR through surveillance networks such as the WHO Global Gonococcal Antimicrobial Surveillance Programme (WHO GASP)[15,153], or the European GASP (Euro-GASP)[154-156] and the U.S. CDC Gonococcal Isolate Surveillance Project (GISP)[157,158]; Euro-GASP and GISP additionally collect clinical and epidemiological data on the corresponding patients.

[H2] Practical applications

Gonorrhoea remains a global public health threat. The biological characteristics of *N. gonorrhoeae* and its proven propensity to develop AMR, the varied clinical manifestations of the infection that may not be obvious or pathogen-specific (particularly for women and extragenital infections), and the limited resources that are dedicated to gonorrhoea control all contribute to the limited success of present gonorrhoea control efforts. Therapy may be hindered by the lack of recommended, high-quality antimicrobials. Current main reliance on only one consistently effective antimicrobial (injectable ceftriaxone) may make effective treatment difficult. Perceptions by patients that they may be resistant or allergic to β-lactam antimicrobials, including ceftriaxone, the logistical constraints of parenteral therapy and fear/avoidance of injections may result in the use of less effective oral therapy. Therapy is also limited in some regions by suboptimal or complete absence of surveillance of infection and particularly AMR, leading to treatment with antimicrobials that will be ineffective due to AMR. Although improved surveillance has increased appreciation of the threat of AMR, this surveillance is not fully representative, being most insufficient or even lacking in areas where the infection is most common[15,153,159].

On the policy level, limited health-care resources directed towards this public health problem (in low-income and middle-income nations and even in high-income nations) have created a tension between diagnostic test cost and assuring a ready supply of medications for gonorrhoea control. The cost of paying for diagnostic testing may erode the funds available for therapy, thereby forcing public health officials to prioritize screening initiatives. In recent years, clinical microscopy (Gram stain) as a low cost POCT has become less available as well, owing to lack of availability of microscopes and adequate technical training in the methodology.

All these challenges are sometimes amplified by social factors. Stigma is a pervasive and powerful force that affects the prioritization of gonorrhoea as a public health problem and influences the behaviour of persons with, or at risk for, gonorrhoea, with regard to health-seeking behaviour and partner notification. Stigma also affects health-care provider attitudes and practices, including evaluation of STI risk and appropriate screening[159].

At the individual level, few persons wish to identify themselves as being at risk for STIs, potentially inhibiting discussion of STI risk with their health care provider, prevention measures, and seeking evaluation for genitourinary symptoms and signs. Limited access to health care may also prevent or delay recommended STI screening or evaluation of symptoms when present. Finally, persons diagnosed with gonorrhoea or other STIs may fail to notify their sex partners of their risk for infection, thereby increasing the probability of complications or continuing transmission.

Clinicians too are sometimes hindered by perceived social factors in evaluating and managing persons with or at risk for STIs. Busy clinicians may assume that their patients are not at risk or hesitate to take sexual histories without a cue to action from their patients, such as a history of possible exposure or genitourinary symptoms or signs, worrying that to ask such questions might be offensive to patients, when data in fact indicate that, if properly presented, this is not the case[159]. Clinician reticence, along with individual embarrassment and/or shame may also hinder partner notification.

Thus while the principles of gonorrhoea management are well known, there are numerous areas within the current management strategies that need to be improved.

[H1] Quality of life

As gonorrhoea is an STI, its diagnosis is often associated with perceptions of social stigma, shame and denial, and can lead to intense embarrassment and fear of retaliation, domestic violence or loss of relationships, including marriages[160]. In the 1960s, the sociologist Erving Goffman described stigma as "undesired differentness" and "discrediting"[161] – a finding reinforced by research findings in the 1990s showing that STI-related stigma resulted in lower testing rates for gonorrhoea[162]. More recent studies have shown that stigma in different populations contributes to a reduction in seeking testing for STIs, reluctance to notify sexual partners and lower levels of treatment compliance[163,164]. For example, in Bhutan, perceived stigma was identified as a key reason for high levels (>50%) of loss to follow-up among patients diagnosed with gonorrhoea[165]. Research found that common coping strategies among people with gonorrhoea in an urban American setting included denial and disengagement–although these behaviours did not affect greatly rates of partner notification[166]. These findings, specific to gonorrhoea, are illustrative of more general findings that stigma influences STI care-seeking. Research noted a reluctance to seek STI testing in young women from socio-economically marginalised neighbourhoods in Canada, owing to "stigma and the fear of being ostracized"[167], and studies found that among African American men increasing STI-related stigma was "significantly associated with…decreased odds of having been tested, [and]…decreased willingness to notify non-main partners"[168]; these factors may contribute to the observed disparities in the distribution of STIs across the intersectional inequalities of ethnicity and gender[169]. In Tigray, Ethiopia, rates of loss to follow up were lower among patients with low levels of STI-related stigma than in study participants reporting high levels of stigma[164].

At the policy level, stigma around gonorrhoea probably contributes to the widespread lack of attention and resource allocation within public health global and national programmes. A recent review of the challenges and opportunities for STI control argued that stigma associated with gonorrhoea and other STIs arises, in part, from "condemnatory moral attitudes" around the behaviours leading to risk of infection – in particular same-sex relationships and transactional sex[170]. Earlier research investigating gonorrhoea control in the USA in the 1970s and 1980s similarly argued that "society's propensity to view gonorrhoea as a disease of "immoral" people" directly contributed to the lack of resources and attention paid to the infection[171]. Qualitative research on the lack of political prioritization afforded to STI control in China confirmed that STIs received a lower place on the health agenda than HIV infection, as decision-makers associated them with "immorality" and patients were considered "condemnable"[172].

It can be argued that the high levels of stigma and accompanying negative framing of gonorrhoea and other STIs exert the most substantial effect on quality of life measures associated with gonorrhoea. Perceptions of embarrassment and humiliation that a diagnosis may bring – both for the affected individuals and their sexual partners – combined with under-resourced public health control programmes, contribute to undiagnosed or poorly treated infections, thereby increasing risks of onward transmission and individual clinical complications and longer-term sequelae caused by this otherwise treatable infection.

Paradoxically, the rise of AMR in *N. gonorrhoeae* may, potentially, force policy-level decision makers to act to devote more attention to the prevention and control of gonorrhoea. However, it should be emphasised that interventions to tackle gonococcal AMR are only likely to succeed if they address not only questions of appropriate antimicrobial use/misuse, but also aim to decrease the global burden of gonorrhoea, which also requires reducing the perception of associated shame and stigma. Effective interventions to decrease stigma and increase patient quality of life should be directed not only at individual and community levels, but also at the political level, to identify and address the social conditions giving rise to stigma and promote institutional fairness[173].

[H1] Outlook

It is imperative to address many global issues for the successful management and control of gonorrhoea. These key priorities and research efforts span all fields, from epidemiology of the pathogen and the disease to the quality of life of patients (Box 2). Of note, reducing the perception of shame, humiliation and stigma that is associated with a diagnosis of gonorrhoea and with certain sexual orientations (for example, MSM) in many settings is crucial to obtain more accurate incidence and prevalence data and to decrease the global burden of gonorrhoea, which would also substantially reduce the gonococcal AMR levels worldwide. Effective interventions to decrease STI-associated stigma should be implemented at individual and community levels, and at the social and political levels where social conditions giving rise to stigma should be identified and tackled[173]. Gonorrhoea and other STIs need to be considered and managed by individuals, the health system, general community, and at political level in all countries in recognition of the right to health services free of discrimination and without stigma.

[H2] Epidemiology

The incidence of gonorrhoea is increasing, especially in high-income settings globally. However, global population-based incidence and prevalence data are extremely scarce from most settings and, even in high-income settings, where surveillance is conducted in a more systematic and regular manner, the surveillance data should be interpreted with caution as the surveillance systems, diagnostic testing, methodologies, and quality assurance are not standardized across countries and remain weak in several settings[33,36]. Additionally, the current prevalence of serious complications and sequelae due to gonorrhoea is mainly unknown and estimates are mostly based on historical data. WGS will revolutionize our understanding of the molecular epidemiology (that is, the geographical and temporal spread) of *N. gonorrhoeae* strains. WGS is substantially more accurate than previously used molecular epidemiological typing methods and can adequately describe the emergence, transmission and evolution of AMR gonococcal strains both geographically and temporally, as well as predict AMR with adequate accuracy[45,156,174-184]. However, it is important to strongly emphasize that the full benefits of using WGS for both molecular and infection epidemiology can only be achieved if the WGS data are linked to phenotypical data for the gonococcal isolates and the clinical and epidemiological data for the corresponding patients with gonorrhoea. Notably, WGS of gonococcal isolates with joint analysis of clinical and epidemiological data has also already been introduced and provided increased understanding of, for example, the distribution of AMR and susceptible gonococcal strains in different populations nationally and regionally in the international Euro-GASP (which currently includes 27 European countries)[156].

[H2] Mechanisms

Our understanding of the pathophysiology of gonorrhoea is still limited in many areas, especially the natural course of the infection (including duration and spontaneous resolution), the dynamics of pathogenesis and infection (such as transmission, average time to detection and treatment in different populations, effects of treatment (or cotreatment for other concomitant STIs) on innate and adaptive immunity, host damage and possible host protection) and immune responses and their suppression in urogenital and particularly extragenital sites, such as the pharynx. Improving the knowledge in these areas would enable to more effectively utilise mathematical modeling in the gonorrhoea and gonococcal AMR field, taking into account microbiological, genomic, evolutionary, clinical immunological, and epidemiological data[185], as well as in vaccine development.

After the introduction of any new therapeutic antimicrobial for gonorrhoea, *N. gonorrhoeae* has rapidly acquired or developed decreased susceptibility or resistance to it (Figure 1) via several AMR mechanisms: enzymatic destruction or modification of the antimicrobial, modification or protection of antimicrobial targets to avoid binding, increased export of the antimicrobial (for example, through the MtrC–MtrD–MtrE efflux pump) and decreased uptake of the antimicrobial (for instance, through the porin PorB)[16]. Some AMR determinants, particularly target alterations, directly cause AMR, whereas others cannot result in AMR on their own and require the presence of additional AMR determinants. The accumulation of many AMR determinants does not appear to substantially reduce the biological fitness of *N. gonorrhoeae*[16-21], and some AMR determinants seem to even enhance the fitness of specific gonococcal strains[19-21]. Nevertheless, we need to substantially improve our understanding and definition of fitness as well as of compensatory mutations that could restore possible fitness cost in *N. gonorrhoeae*. We need detailed knowledge regarding how gonococcal AMR determinants affect the fitness of gonococcal strains, how fitness affects the emergence and spread of AMR strains and how these strains become established in the circulating gonococcal populations. Thus, we need to investigate how the fitness of AMR strains may affect the competition with wild type antimicrobial susceptible strains (which is mainly the current fitness definition in microbiological research) and its effects on several factors, such as transmissibility, duration of infection in different anatomical sites, and proportion of symptomatic and asymptomatic infections and severe complications and sequelae in heterogeneous populations with different sexual behaviours. Further research is also needed to identify and characterize in detail known or novel AMR determinants in clinical gonococcal isolates (including their induction and selection, evolution, effect on AMR and biological fitness), and to develop and evaluate genetic AMR prediction tests that can supplement the culture-based AMR surveillance.

[H2] Diagnosis, screening and prevention

In many settings, mostly in less-resourced areas (in which frequently the prevalence of gonorrhoea is the highest), the diagnosis, testing, case reporting, and prevention of gonorrhoea remain suboptimal. Thus, it is important to widely implement the use of cost-effective, appropriate, and quality-assured NAATs. If required, these NAATs can be performed in centralized reference laboratories for cost-effectiveness and to maintain a high level of quality assurance. In addition, rapid, appropriate POCTs for the diagnosis of gonorrhoea and other STIs are urgently needed. Gonococcal POCTs should ideally simultaneously predict AMR to inform treatment. For some antimicrobials, such as ciprofloxacin, mathematical modeling has indicated that POCTs with high sensitivity to detect AMR can be more effective than NAATs and even culture to preserve the effectiveness of the antimicrobial. By contrast, POCTs detecting *N. gonorrhoeae* without reliable AMR detection may accelerate the spread of AMR gonococcal strains[186]. Several rapid, sensitive and specific NAAT-based POCTs for gonorrhoea are in the pipeline and will be available the coming few years (Table 3)[101,122,170,187]. Accordingly, it will soon be essential to prepare health care systems for use of these POCTs, by including them in STI training modules, management guidelines, diagnostic algorithms, and regulatory frameworks. Limitations to the adoption of POCTs are considerable and include time for results; cost of the instrument; lack of required infrastructure, quality assurance and reporting criteria; supply chain issues that may discourage use; lack of clear recommendations on the inclusion of POCTs in diagnostic algorithms and regulatory frameworks, lack of training opportunities and education of health care workers about the utility and advantages of POCTs; and worries by laboratory-based personnel that out-of-lab testing may infringe on job security[118].

In an era of high prevalence of AMR in *N. gonorrhoeae* coupled with the widespread use of diagnostic gonococcal NAATs internationally, it is essential to retain and additionally strengthen the ability to perform gonococcal culture, which is the only method that enables complete AMR testing, because surveillance of gonococcal AMR (preferably MIC-based) and ideally also of cases of treatment failure is imperative. In settings where NAATs solely are used for diagnosis of gonorrhoea, participation in organised and quality-assured national, regional and/or international GASPs is crucial.

WGS and other new technologies such as transcriptomics and proteomics are also informing the development of *N. gonorrhoeae* diagnostics and vaccine[156,174-183,188-191]. For developing gonococcal vaccines, a number of promising protein antigens have been described and characterized, including proteins involved in colonisation (for example, PilC, PilQ, PorB, Opa, and OmpA), evasion of innate defenses (for example, MtrE, SliC, Ng-ACP, MsrAB, Lst, and PorB) and nutrient acquisition (for example, TbpA, TbpB, LbpA, and LbpB); structural proteins (for example, BamA, BamE, NGO2054 and NGO2111); other proteins such as AniA (implicated in nitrate reduction) and MetQ (methionine transporter that promotes survival in macrophages); the 2C7 epitope (peptide mimetic of LOS epitope); and OMVs[131,132,134]. Many of the promising new vaccine targets for *N. gonorrhoeae* have been identified through proteomic approaches and transcriptome analysis of genes expressed during gonococcal infections[188-190,192]. Furthermore, to overcome the restrictions of the current model of female mice treated with 17β-estradiol new animal models for *N. gonorrhoeae* infection are being developed, such transgenic mice that mimic human infections and express human cell adhesion molecules or iron binding molecules[193,194], and a transgenic mice model expressing human complement Factor H is available for the closely related *N. meningitidis*[195].

[H2] Management

Currently available genetic assays have shortcomings (such as cross-reactions with non-gonococcal *Neisseria* species in clinical, particularly pharyngeal, specimens, and suboptimal sensitivity and/or specificity) that limit their prediction of resistance or susceptibility to currently recommended therapeutic antimicrobials (except for ciprofloxacin, for which the sensitivity and specificity of NAATs are generally >95%), and newly emerging AMR determinants are not detected[196-198]. However, future improved rapid POCTs that detect both *N. gonorrhoeae* and its resistance or susceptibility to several antimicrobials will guide individualized therapy at the first health-care visit and restrict the use of last-line antimicrobials[196-199]. Such POCTs will improve the management and control of both gonorrhoea and *N. gonorrhoeae* AMR. WGS can also be utilised for prediction of AMR and MICs of antimicrobials with reasonably high accuracy[156,182-184]. Rapid, real-time sequencing with the hand-held MinION sequencer was shown to generate fairly accurate genome sequences and be able to predict resistance to ciprofloxacin and azithromycin and decreased susceptibility or resistance to cefixime in *N. gonorrhoeae*[183]. The rapid development of WGS technologies with decreasing complexity and cost and faster turnaround times may make these technologies suitable for *N. gonorrhoeae* detection and prediction of resistance or susceptibility to therapeutic antimicrobials at the diagnostic setting, including at POC.

The global issue of AMR in *N. gonorrhoeae* will probably continue to escalate and we cannot rely on the last-line ceftriaxone (plus azithromycin) indefinitely. Consequently, new antimicrobials, with novel mechanisms of action, for monotherapy and/or inclusion in dual therapies for urogenital and extragenital gonorrhoea are crucially needed. Some recently developed new antimicrobials, the spiropyrimidinetrione zoliflodacin[200-204], and triazaacenaphthylene gepotidacin[205-207], will both soon enter Phase 3 randomised clinical controlled trials for uncomplicated gonorrhoea. Additional promising novel antimicrobials in earlier development that deserve further attention for treatment of gonorrhoea (and possibly additional STIs) are, for example, lefamulin[208,209] and SMT-571[210]. However, until novel antimicrobials are available, it is imperative to increase our knowledge regarding ideal treatment, including dosing regimens, of gonorrhoea and other STIs, such as *C. trachomatis* and *M. genitalium* infections, with the available antimicrobials ceftriaxone, azithromycin and doxycycline. Clearly, a more holistic view on the treatment of bacterial STIs and understanding the effect of any new bacterial STI treatment on other STI pathogens and the bystander microbiota is essential. Current knowledge regarding the pharmacokinetics and pharmacodynamics of the available antimicrobials in the treatment of gonorrhoea and other STIs at urogenital and particularly extragenital sites is highly limited[211] and requires substantially increased attention to inform ideal dosing regimens, and multiple dose regimens for gonorrhoea might be required.

Box 1. ***Models to study N. gonorrhoeae pathogenesis***

Much of the information concerning *N. gonorrhoeae* pathogenesis has come from studying the physiological and genetic properties of the organism, including determination of growth and nutrient requirements and surface-exposed molecules, with *in vitro* bacterial cultures. However, these experimental conditions do not always mirror *in vivo* conditions, and, therefore, cell culture models can be useful to learn about the interactions between the bacterium and the host, particularly how *N. gonorrhoeae* attaches to and is internalized into eukaryotic cells. These studies have mainly used immortalized transformed human cell lines, but occasionally utilized newly-harvested human primary cells[1], as cell lines do not always replicate the properties of tissues. Primary cultures are difficult to isolate and maintain and are substantially heterogeneous, whereas tissue explants enable to study the interactions of the organism with different cell types in a complex tissue. Compared with other primary tissues, fallopian tube tissue is relatively easy to obtain from hysterectomies and is a clinically relevant tissue environment[70], particularly for modelling PID[212].

Animal models are useful to study colonisation, growth and immune response in a host. Of note, because *N. gonorrhoeae* is restricted to the human host, the bacterial proteins have evolved highly specific interactions with human molecules, rendering early mouse models of limited value. Despite this limitation, female mice treated with 17β-estradiol (to promote prolonged colonization and/or infection) have become a standard in the field[193]. Transgenic mouse models expressing human receptors for *N. gonorrhoeae* are in development and will have greater utility in the future[194,195], although no existing mouse model totally mimics a natural human infection. In the 1960s, primate models were examined and chimpanzees reportedly developed symptomatic gonorrhoea[213], but chimpanzees are no longer used for biomedical research in the USA and rarely elsewhere, although new primate models might be developed in the future. The human challenge model is the most relevant existing model[214]. Only men can participate, as they have lower risk for complications of infection than women. This model has only been used to investigate initial colonisation determinants, and its utility is limited owing to small cohorts per study, the requirement for treatment as soon as symptoms develop, and being applicable to men only.

Box 2: Key priorities in gonorrhoea research and control

- Decreasing the perception of stigma, humiliation and shame associated with gonorrhoea and other STIs, and ensuring that services and interventions are delivered free of discrimination, leaving no populations behind

[H1] Epidemiology

- Increasing knowledge of the incidence and prevalence of the infection and its complications and sequelae in general population and subpopulations
- Expanding global AMR surveillance (phenotypic and genetic AMR testing), including surveillance of treatment failures and antimicrobial use/misuse, in combination with whole genome sequencing and clinical and epidemiological data of patients

[H1] Mechanisms/pathophysiology

- Improving knowledge of natural course and pathogenesis, including genomic, physiological and pathogenic/virulence mechanisms of *N. gonorrhoeae*, in different anatomical sites and understanding the emergence, evolution, spread, and biological costs or benefits (fitness) of AMR
- Understanding of pharmacokinetics and pharmacodynamics of current and future therapeutic antimicrobials in urogenital and particularly extragenital sites, to inform treatment guidelines

[H1] Diagnosis, screening and prevention

- Increasing diagnostic testing (also to detect asymptomatic gonorrhoea), use of validated and quality-assured NAATs, and developing rapid, appropriate, and affordable POCTs, which should also enable simultaneous prediction of antimicrobial resistance or susceptibility status
- Strengthening prevention (for example, increasing the use of condoms and of out-of-box approaches, such as the use of antiseptic mouthwash to prevent acquisition and transmission of pharyngeal gonorrhoea[215])
- Improving the understanding of the effects of PrEP on prevalence of gonorrhoea and other STIs in different populations, the risk factors involved, and the ideal counselling, monitoring and screening intervals for individuals taking PrEP
- Developing gonococcal vaccine(s), for which substantial progress has been made in recent years[131,132,134,136-138,189,191,216]

[H1] Management

- Promoting early diagnosis and treatment of patients and their partners, following evidence-based international and national guidelines
- Promoting responsible antimicrobial use and stewardship (both STI-related and on a population level), as excessive antimicrobial use can decrease the susceptibility of *N. gonorrhoeae* to therapeutic drugs, both directly (through selection of AMR in *N. gonorrhoeae*) and indirectly (through selection of AMR determinants in for example commensal *Neisseria* spp. that are subsequently shared through HGT with *N. gonorrhoeae*[217])
- Developing novel therapeutic antimicrobials and strategies to preserve the efficacy of current and future antimicrobials

Figure 1 Recommended empiric therapy for gonorrhoea and emergence of antimicrobial resistance in *Neisseria gonorrhoeae*

Each bar represents a gonorrhoea therapy, and the length of the bar represents the time period from when the therapy started to be used until when clinical and/or *in vitro* resistance threatening the efficacy of that specific antimicrobial therapy had emerged. *In vitro* verified antimicrobial resistance (AMR) determinants are also shown[16-21,218-220]. PBP2 amino acid alterations that increase the minimum inhibitory concentration (MIC) of extended-spectrum cephalosporins (ESCs) (verified, for example, by site-directed mutagenesis or transformation) in non-mosaic and mosaic (in which concomitant epistatic mosaic *penA* mutations are also needed) *penA* alleles are noted by an asterisk[218-220]. Additionally, PBP2 G542S, P551S, and P551L amino acid alterations in non-mosaic *penA* alleles have been statistically associated with gonococcal strains with decreased susceptibility to ESCs[221-223]. It is a grave concern that during the past decade(s) resistance to azithromycin and decreased susceptibility to the ESC ceftriaxone, the last remaining option for empirical monotherapy, have been reported worldwide. The first *Neisseria gonorrhoeae* strain with high-level resistance to ceftriaxone was isolated in 2009 in Japan, which was followed by some isolates with high-level ceftriaxone resistance in 2011 in France and Spain. During subsequent years, ceftriaxone resistant isolates have been characterised in many countries including Japan, China, Australia, Singapore, Canada, Argentina, and several European countries. Furthermore, treatment failures with ceftriaxone were verified in Japan, Australia, and in several European countries[15,16,153,224-240]. In 2014, the first failure of ceftriaxone–azithromycin dual therapy for gonorrhoea was verified in the UK[241]. Worryingly, since 2015, an international spread of one ceftriaxone-resistant gonococcal strain, initially described in Japan, has been confirmed[229-235,239,240,242,243], and the first strain with resistance to ceftriaxone plus high-level azithromycin resistance was isolated in 2018 in the UK and Australia[236-238].
AZM, azithromycin; CFM, cefixime; CRO, ceftriaxone; DOX, doxycycline.

Figure 2 Estimated new global cases of gonorrhoea in 2016

Estimated numbers (in millions) of incident cases of gonorrhoea in adults (15–49 years of age), by WHO region[22]. These data correspond to 20 new gonococcal infections per 1,000 women and 26 per 1,000 men globally. The highest incidence rates were in the WHO African region, with 41 cases per 1,000 women and 50 per 1,000 men, followed by the WHO Region of the Americas, with 23 cases per 1,000 women and 32 per 1,000 men; the lowest incidence was in the WHO European Region, with 7 cases per 1,000 women and 11 per 1,000 men[22]. The World Bank Income Classification (https://databank.worldbank.org/reports.aspx?source=2&series=NY.GNP.PCAP.CD&country=) is also shown. Permission lines required (data from)

Figure 3 *Neisseria gonorrhoeae* cell envelope structure

Neisseria gonorrhoeae is a Gram-negative bacterium, frequently encountered as diplococci (individual cells are ~0.6–1 µm in diameter), with a characteristic cell envelope consisting of a cytoplasma membrane (the inner membrane), a periplasmic space containing the peptidoglycan cell wall[244] and the outer membrane containing lipooligosaccharide (LOS), which is similar to lipopolysaccharide (LPS) of other Gram-negative bacteria, except it does not have the polymeric O-antigen characteristic of LPS. The Type IV pilus is a long, thin fiber that reaches far outside of the cell envelope, mainly composed of many copies of one protein, pilin. Type IV pilus assembly requires a complex molecular machine, called the assembly apparatus, that sits within the cell envelope to produce the fiber on the outside of the cell[245]. The pilus is a dynamic structure that can be retracted by the assembly apparatus, which generates one of the largest physical forces on record by a biological machine[246]. The Opa proteins are a family of integral outer membrane proteins whose expression is stochastically controlled[247]. Each *N. gonorrhoeae* isolate carries ~11 *opa* genes, and expression of each is controlled by independent molecular events that turn on or off the expression of each *opa* gene. A single bacterial cell may express none of the Opa proteins, a single Opa, or a combination of several. There is a correlation between patterns of Opa expression and bacteria isolated from females during menses[248], and increased numbers of Opa proteins are expressed during human volunteer infections[249]. The outer membrane localized porin (PorB) allows small molecules to enter the periplasm and the reduction modifiable protein (Rmp) is associated with PorB and elicits antibodies that block the binding of anti-PorB antibodies[250]. The three iron scavenging complexes (LpbA–LpbB, HpuA–HpuB, and TbpA–TbpB) are required to obtain iron from the host. Adapted from Ref[5]. Permission lines required

Figure 4 *Neisseria gonorrhoeae* infection
Initial adhesion of *Neisseria gonorrhoeae* to the epithelium requires type IV pili and then Opa proteins for more intimate adhesion. The bacteria can then proliferate on the epithelial surface and invade underlying tissues via transcytosis. *N. gonorrhoeae* also releases peptidoglycan fragments, OMVs and LOS, thereby activating Toll-like receptors (TLRs) and nucleotide-binding oligomerization domain-containing protein (NOD) signalling in tissue resident dendritic cells and macrophages. In response to bacterial stimulation, these cells produce chemokines and cytokines (for example, IL-1, IL-6, IL-8, IL-17 and tumor necrosis factor (TNF)) that can recruit polymorphonuclear leukocytes (PMNLs); however, the bacteria can often survive phagocytosis, antibacterial factors released during degranulation, or NETosis. *N. gonorrhoeae* has many ways to prevent complement killing by the membrane attack complex; for example, the LOS can be modified by sialic acid, when the precursor substrate, CMP-NANA, is supplied by the host, to enhance complement resistance[251]. Sialylated LOS binds C3b and promotes its inactivation to iC3b via factor I, whereas PorB binds factor H and C4BP, thereby hiding the bacteria from complement recognition. When complement activity is inhibited (for example by mutation or owing to immune suppressive treatment), systemic *N. gonorrhoeae* infections are prevalent. It is not known if the resistance to complement is also important in localized sites of colonization. Adapted from Ref[5]. Permission lines required.

Table 1 **Tests for the diagnosis of *Neisseria gonorrhoeae*[a]**

Parameter		**Microscopy**	**Culture**	**NAAT**
Specimen types[b]				
Endocervical swab		Yes[c]	Yes	Yes
Vaginal swab		No	Yes[c]	Yes
Urine	*Female*	No	No	Yes[c]
	Male	No	No	Yes

Urethral swab	Yes	Yes	Yes
Rectal swab	No	Yes	Yes/No[d]
Pharyngeal swab	No	Yes	Yes/No[d]
Conjunctival swab	Yes	Yes	Yes/No[d]
Performance			
Sensitivity[e]	Low–high	Moderate–high	Very high
Specificity[e]	Moderate–high	Very high	Moderate-very high
Cost	Low	Moderate	Moderate-very high
Instrumentation	Microscope	Routine microbiology	Moderate-large footprint
Technical complexity	Low-moderate	Moderate	Low-High
Level of laboratory infrastructure	Low	Low–intermediate	Intermediate–high
Potential as a POCT	Yes	No	Yes

a) Modified from Ref[85]
b) Yes or No indicates appropriateness of specimen type.
c) The sensitivity is substantially lower than in other approved specimen types and a negative result does not exclude gonococcal infection.
d) Yes/No indicates that not all platforms have received FDA approval for that specific specimen.
e) Can highly depend on specimen type.

Table 2 **US FDA-approved and CE-IVD-approved NAATs for the detection of *Neisseria gonorrhoeae***

Test, instrument (manufacturer)	**Gonococcal target(s)**	**Specimen type or cultured isolates**	**Sensitivity (%)**		**Specificity (%)**		**References**
			Symptomatic	**Asymptomatic**	**Symptomatic**	**Asymptomatic**	
PCR							
RealTime CT/NG[a], m2000[b] (Abbott Molecular)	*opa*	CCVS (F)	96.8	95.7	99.9	99.4	[107,252,253]
		ECS (F)	87.1	91.3	99.7	100	
		FVU (F)	76.9	NA	99.8	NA	
		SCVS (F)	96.7-98	95.7	99.7-100	100	
		Urethral (M)	99.2	81.8	99.3	99.8	
		Urine (F)	93.8	87	99.7	99.6	
		Urine (M)	98.8	100	99.5	100	
		Urine (M/F)	100	NA	100	NA	
		Culture	99.5	NA	100	NA	[100]
Xpert CT/NG[a,c,d], GeneXpert (Cepheid)[b]	Two (NG2, NG4) highly conserved, noncontiguous unique chromosomal targets	ECS (F)	100	100	100	100	[254]
		Urine (F)	100	91.7	100	99.9	
		Urine (M)	97.8	100	100	99.9	
		VS (F)	100	100	99.8	99.9	
		Culture	100	NA	100	NA	[187,255]
Cobas 4800 CT/NG[a], Cobas 4800[b] (Roche)	Direct Repeat Region 9 (DR9)	FVU (F)	81.1	NA	100	NA	[107 256,257]
		Nongenital (F)	100	NA	100	NA	
		Nongenital (M)	100	NA	99.8	NA	
		SCVS (F)	84.6	NA	99.6	NA	
		Urine (M/F)	92.9	NA	100	NA	
		Urogenital (F)	97.5	NA	100	NA	
		Urogenital (M)	100	NA	100	NA	
		Culture	100	NA	100	NA	[100]
BD MAX[e], BD	Chromosomal	ECS	96.3	94.1	99.9	100	[258]

Max System (Becton-Dickinson)	DNA	Urine (F)	100	88.9	99.9	99.5	
		Urine (M)	NA	80	NA	100	
		VS	96.3	94.1	99.8	99.9	
SDA							
Probe Tec ET[a], Viper XTR (Becton-Dickinson)	Pilin gene-inverting protein homologue	ECS (F)	87.5	91.3	99.6	98.9	107,252,253,259
		FVU (F)	75.5	NA	100	NA	
		SCVS (F)	90.6-100	NA	100	NA	
		Urine (F)	76.7	85.7	95.6	96.9	
		Urine (M)	94.9	100	97	95.7	
		Urine (M/F)	95.8	NA	100	NA	
		CCRS (M)	67.5	NA	100	NA	106,260
		OPS (M)	85.7	NA	100	NA	
		Rectal (M)	89.1	NA	99.8	NA	
		SCRS (M)	77.1	NA	99.3	NA	
		Culture	100	NA	88.9	NA	100
TMA							
Aptima Combo 2[a,d], Panther[b] (Hologic (earlier Gen-Probe))	16S rRNA	CCVS (F)	93.8	95.7	99.3	99.7	107,252,253,259
		ECS (F)	90.6	90.9	99.4	99.7	
		FVU (F)	88	NA	99.4	NA	
		SCVS (F)	96.2-100	NA	98.4-100	NA	
		Urethral (M)	99.2	81.8	99.2	99.7	
		Urine (F)	84.4	82.6	99.6	99.4	
		Urine (M)	97.9	100	99.7	99.5	
		Urine (M/F)	100	NA	100	NA	
		CCRS (M)	78.3	NA	99.8	NA	106,260
		OPS (M)	100	NA	99.6	NA	
		Rectal (M)	93.5	NA	97.7	NA	
		SCRS (M)	84.3	NA	100	NA	
		Culture	100	NA	100	NA	100,261

All tests are both approved by the US FDA and have a CE (European Conformity) – IVD (in vitro diagnostic) certification, indicating compliance with health, safety, and environmental protection standards for products manufactured or sold within the EU/EEA[101]. A large number of additional NAATs (not shown) carry only a CE-IVD certification, in general, these NAATs are less stringently validated.

a) Can also detect *Chlamydia trachomatis*
b) Fully automated.
c) Cartridge-based near-POCT
d) FDA approved for extragenital specimens such as rectal and pharyngeal infection[105]
e) Can also detect *C. trachomatis* and *Trichomonas vaginalis*

PCR, polymerase chain reaction; SDA, strand displacement amplification; TMA, transcription mediated amplification. SCVS, self-collected vaginal swabs; FVU, first void urine; CCVS, clinician-collected vaginal swabs; VS, vaginal swabs; ECS, endocervical swab; SCRS, self-collected rectal swab; CCRS, clinician-collected rectal swab; OPS, pharyngeal swabs; F, female; M, male; NA, Not available in the referenced studies.

Table 3 **POCTs, near-POCTs and antimicrobial resistance tests available and in pipeline[a]**

Platform/Test	GeneXpert Xpert CT/NG[b]	Binx io CT/NG	ID NOW CT/NG[c]	Truenat CT/NG	ResistancePlus GC[d]
Manufacturer	Cepheid	Atlas Genetics	Abbott	Molbio	SpeeDx
Instrument; healthcare setting	Table-top, not portable (used in mobile clinics); Level 2	Table-top, portable; Level 1	Table-top, portable; Level 1	Table-top, portable; Level 2	Table-top PCR machines, not portable; Level 2
Amplification technology	PCR	NAAT, immunoassay and small molecule chemistry	Isothermal PCR	Real-time PCR	Real-time PCR
Specimen	Female and male urine, endocervical swab and patient-collected vaginal swab	Self-collected and clinician-collected vaginal swabs from symptomatic and asymptomatic females, and urine from males.	TBD	Endocervical and vaginal swabs, male urethral swab, male and female urine	Male and female urine; rectal, cervical, vaginal, urethral, pharyngeal, and ocular swabs; and ocular extracts
Procedure	~4 steps, sample preparation automated	~4 steps, sample preparation automated	~6 steps, raw sample added to device	Multiple pipetting steps	~4 steps
Time to result	~90 minutes	30 minutes	15 minutes	~60 minutes	50 minutes
Reagent stability	3 years	Cartridges with reagents stable at 2-25°C	>12 months	2 years at temperatures 2-30°C	18-24 months
Energy requirements	Mains power required; solar power possible, can be powered by 12V DC or 120V AC	Mains power required	AC mains and DC from external AC/DC supplied plug pack	Rechargeable lithium ion battery	Mains power required
Training	Less than ½ day	Less than 1 hour	Less than ½ day	Less than ½ day	Less than ½ day
Connectivity	Yes, computer required, remote calibration	Yes, via middleware	Yes, USB and Ethernet outlets	Yes, wireless connectivity: Wi-Fi, Bluetooth, SMS	Yes, computer required
Regulatory	FDA, CE-IVD	CE-IVD, FDA	N/A	CE-IVD	CE-IVD, FDA

Compliance		approval pending		approval pending	approval pending

PCR, polymerase chain reaction: NAAT, nucleic acid amplification test; N/A, Not available; Level 1 – primary healthcare center; Level 2 – district hospital; TBD, to be determined.

a) This table is not an exhaustive list of all POCTs in the pipeline; the tests listed were selected because there is more information available[101,170].
b) Near-POCT
c) Previously named Alere i CT/NG.
d) First licensed molecular test detecting both *N. gonorrhoeae* and its ciprofloxacin susceptibility status[101,170]

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