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212 *Neisseria gonorrhoeae* (Gonorrhea)

Jeanne M. Marrazzo and Michael A. Apicella

SHORT VIEW SUMMARY

Definition
- Gonorrhea is a sexually transmitted infection caused by the organism *Neisseria gonorrhoeae.*

Epidemiology
- Gonorrhea occurs most commonly in adolescents and young adults worldwide.
- Overall, incidence has been declining; however, an exception is among men who have sex with men.
- *N. gonorrhoeae* should be considered in the evaluation of genital inflammatory syndromes, including cervicitis, urethritis, and pelvic inflammatory disease.
- Most infections, however, involve neither signs nor symptoms; thus, routine screening of young women (≤25 years) or older women with key risk factors is recommended.
- Untreated maternal infection may result in ophthalmia neonatorum, which may be avoided with neonatal prophylaxis.

Microbiology
- This gram-negative, intracellular diplococcus is a fastidious microorganism that grows only in vitro in a narrow temperature range (35°C–38°C) on complex media.
- It has a marked ability to develop resistance to antibiotics; recently there has been the global appearance of strains with increasing resistance to third-generation cephalosporins.
- The gonococcus is naturally competent. This is a factor that has led to the rapid acquisition of antimicrobial resistance.
- *N. gonorrhoeae* is an exclusively human pathogen. It uses different pathogenic mechanisms to infect the epithelium of men and women. In men, the asialoglycoprotein receptor present on the urethral epithelia interacts with the terminal galactose of the lipooligosaccharide (LOS) to enter human cells. In women, a cooperative interaction occurs among C3b covalently linked to the LOS, gonococcal porin and pilus, and complement receptor 3 (CR3) on the surface of the cervical epithelial cell.

Diagnosis
- Culture and nucleic acid amplification testing are sensitive and specific and can be performed at all potentially infected anatomic sites.

Therapy
- Options are limited, given widespread resistance to numerous antibiotic classes.
- Parenteral therapy with a third-generation cephalosporin is currently recommended and should be accompanied by treatment with azithromycin for additional coverage (see Table 212.2).
- Sex partners of infected people should be treated presumptively, regardless of diagnostic test results.
- Patients in whom treatment with standard regimens fails should undergo assessment for reinfection; if reinfection is unlikely, antibiotic resistance should be considered.

Prevention
- Because of the asymptomatic nature of most infections, routine screening of young women (≤25 years) and pregnant women is recommended.
- Condoms are very effective in preventing transmission.

Gonorrhea is a common bacterial infection that is transmitted almost exclusively by sexual contact or perinatally and primarily affects the mucous membranes of the urethra and cervix and, less frequently, those of the rectum, oropharynx, and conjunctivae. Ascending genital infection in women leads to endometritis and salpingitis—collectively called pelvic inflammatory disease (PID)—the predominant complication and one of the most common causes of female infertility. Other complications include acute epididymitis; ophthalmitis; disseminated infection with arthritis, dermatitis, and sometimes endocarditis; and transmission to the neonate with attendant conjunctivitis (ophthalmia neonatorum).

Gonorrhea is one of the oldest known human illnesses, and references to sexually acquired urethritis can be found in ancient Chinese writings, the biblical Old Testament (Leviticus), and other works of antiquity. Galen (AD 130) introduced the term *gonorrhea* ("flow of seed"), implying interpretation of urethral exudate as semen. The causative organism was described by Neisser in 1879 and was first cultivated in 1882 by Leistikow and Löffler. Untreated infections were understood to resolve spontaneously over several weeks or months, but reinfection was recognized. Many therapies were tried, but not until the advent of the sulfonamides in the 1930s and penicillin in 1943 was truly effective treatment available. Since that time, therapy has changed radically in the face of antimicrobial resistance. We are now facing a major crisis as resistance to the last of the "single-dose" third-generation cephalosporin therapies is increasing. Growth of fundamental knowledge about the organism and the host response to infection was slow for 80 years, but a surge of new information began in the 1970s, and the molecular biology of the gonococcus and the pathogenesis of gonorrhea have been well elucidated. Public health control efforts have met with variable success, and gonorrhea remains the second most common reportable disease in the United States (following sexually transmitted chlamydial infection), a prime example of the influence that social, behavioral, and demographic factors can have on the epidemiology of an infectious disease despite highly effective antimicrobial therapy.

THE ORGANISM

Description

Neisseria gonorrhoeae is a non–spore-forming, gram-negative coccus that characteristically grows in pairs (diplococci) with adjacent sides flattened. It does not have flagella but can move based on extension and retraction of its type IV pilus.[1] This process is known as "twitching" and is the basis of the colony types seen in piliated and nonpiliated gonococcal strains. The gonococcus closely resembles the related pathogen *Neisseria meningitidis* and several species of nonpathogenic *Neisseria*. All *Neisseria* spp. rapidly oxidize dimethyl-*p*-phenylenediamine or tetramethyl-*p*-phenylenediamine, the basis of the oxidase test. Traditionally, gonococci are differentiated from other *Neisseria* by their ability to grow on selective media; to use glucose but not maltose, sucrose, or lactose; and to reduce nitrites, and also by their inability to grow well at reduced temperature or on simple nutrient agar.[2]

Growth and Cultivation

Gonococci do not tolerate drying, and patient samples to be used for cultivation should be inoculated immediately onto an appropriate growth or transport medium. Growth is best for most strains at 35°C to 37°C,

and many freshly isolated strains have a relative or absolute requirement for atmospheric carbon dioxide in concentrations of approximately 5%. All strains are strictly aerobic under usual growth conditions, but the organism grows anaerobically when nitrite is provided as an electron acceptor. Colonies appear in 24 to 48 hours, but on most media viability is rapidly lost after 48 hours because of autolysis.[2]

Gonococci are inhibited by many fatty acids, and it is necessary to incorporate starch or other substances that absorb fatty acids into most growth media. All strains have complex growth requirements, including requirements for several vitamins, amino acids, iron, and other factors. For clinical purposes, a satisfactory medium is chocolate agar enriched with glucose and other defined supplements. Isolation of gonococci from sites that normally contain high concentrations of the normal microbiota, especially the pharynx, rectum, and cervix, may be difficult because of overgrowth of the hardier normal microbiota—a problem that is largely overcome with use of media containing antimicrobial agents that inhibit most nonpathogenic *Neisseria* and other species but permit growth of most strains of *N. gonorrhoeae* and *N. meningitidis*. Chocolate agar that contains vancomycin, colistin, nystatin, and trimethoprim (modified Thayer-Martin medium) is widely used for this purpose in the United States; a similarly constituted translucent selective medium (New York City medium) is also commonly used. Specimens from sites that usually do not harbor indigenous microbiota (e.g., blood, synovial fluid, and cerebrospinal fluid) should be cultured on antibiotic-free media.

Surface Structures

The envelope of *N. gonorrhoeae* is similar in basic structure to that of other gram-negative bacteria. As the interface between the gonococcus and host, the cell surface has been intensively studied (Fig. 212.1), and specific surface components have been related to adherence, tissue and cellular penetration, cytotoxicity, and evasion of host defenses both systemically and at the mucosal level.

Type IV Pili

Type IV pili (fimbria) are strong flexible filaments extending from the gonococcal cell surface. They can be several microns in length and 50 to 80 angstroms in width. Pili traverse the outer membrane of the gonococcus through an integral outer membrane protein known as PilQ.[3] Mature pili are composed of repeating protein subunits (pilin) with a molecular weight of 19 ± 2.5 kDa.[4] Through the activity of the proteins PilT and possibly PilB, gonococci can depolymerize and repolymerize the pilus strand, causing the bacteria to "twitch." The pilin subunit has regions of considerable interstrain antigenic similarity, especially near the amino terminus, but areas of extreme antigenic variability are also present.[4,5] A single strain of *N. gonorrhoeae* is capable of producing pili with differing antigenic compositions. This has compromised the usefulness of pilus-based vaccines against gonorrhea. The presence or absence of pili result in varied colonial forms, which can be distinguished when *N. gonorrhoeae* is grown on translucent agar.[6] Fresh clinical isolates initially form colony types P^+ and P^{++} (formerly called T1 and T2), and the organisms have numerous pili extending from the cell surface (Fig. 212.2); P^- colonies (formerly T3 and T4) lack pili. Piliated gonococci are better able than organisms from P^- colonies to attach to human mucosal surfaces and are more virulent in animal and organ culture models and in human inoculation experiments than nonpiliated variants. Expression of pili is a function of the *pil* gene complex. A spontaneous shift between P^+ or P^{++} colonies to P^- colony types, known as phase variation, occurs after 20 to 24 hours of growth in vitro. This is caused by errors in DNA replication, resulting in *pilE* genes that are out of frame and fail to produce functional protein.[5]

In addition to mediating attachment, pili contribute to resistance to killing by neutrophils. In the fallopian tube mucosa model (Fig. 212.3), pili facilitate attachment to nonciliated epithelial cells, which

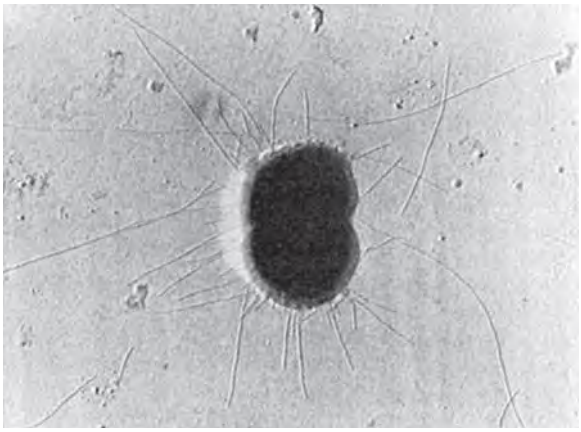


FIG. 212.2 *Neisseria gonorrhoeae* with numerous pili extending from the cell surface. *(Courtesy Dr. Gour Biswas, University of North Carolina School of Medicine, Chapel Hill, NC.)*

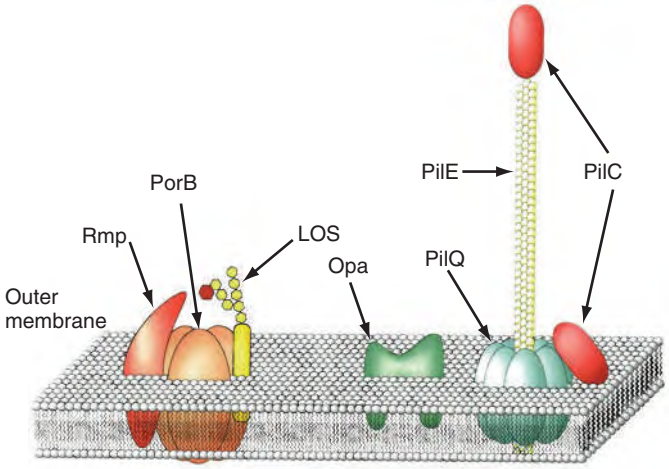


FIG. 212.1 Illustration of gonococcal outer membrane depicting many of the antigens described in the text. *LOS,* Lipooligosaccharide: the branched and phase variable LOS polysaccharide chain is shown as being bound by sialic acid, designated by a red hexagon; *Opa,* opacity protein; *PilC,* the outer membrane protein PilC, which is proposed to be presented by the pilus fibril as a tip adhesin; *PilE,* pilin, the subunits that are assembled into the α-helical pilus fibril; *PilQ,* pilin accessory protein Q, a secretin through which the assembled pilus extrudes; *PorB,* porin protein; *Rmp,* reduction modifiable protein. *(Courtesy C.E. Thomas, University of North Carolina School of Medicine, Chapel Hill, NC.)*

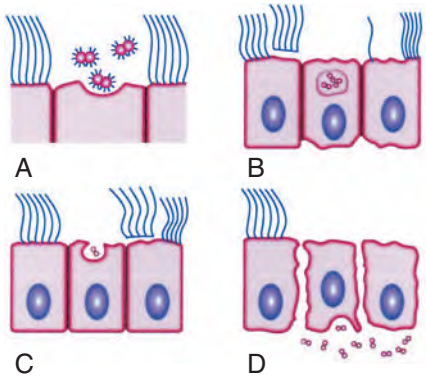


FIG. 212.3 Schematic representation of the interaction between fallopian tube explant epithelial cells and *Neisseria gonorrhoeae.* (A) Attachment of the piliated gonococci to the surface of a nonciliated host cell. (B) Endocytosis of gonococci and loss of cilia on adjacent cells, mediated by lipooligosaccharide (LOS). (C) Transport of gonococci through an epithelial cell in an endocytotic vacuole, in which the organism may replicate; progression of LOS-associated cytotoxicity. (D) Release of organisms into subepithelial space. *(From Dallabetta G, Hook EW III. Gonococcal infections.* Infect Dis Clin North Am. *1987;1:25–54.)*

initiates a process of entry and transport through these cells into intercellular spaces near the basement membrane or directly into the subepithelial space while concurrently nearby ciliated mucosal cells lose their cilia and are sloughed.[7] CD46 was considered to be the main pilin receptor, but the issue is currently uncertain and the identity of the pilus receptor is an area of active study. Other pilus-associated proteins are likely to be important to adhesion to host cells, particularly PilC.[8,9] Other factors also mediate attachment, notably opacity (Opa) proteins, lipooligosaccharide (LOS), and porins (Por).

Outer Membrane

Like all gram-negative bacteria, the gonococcus possesses a cell envelope composed of three distinct layers: an inner cytoplasmic membrane, a middle peptidoglycan cell wall, and an outer membrane. The outer membrane contains LOS, phospholipid, and a variety of proteins (see Fig. 212.1). Porin, formerly designated protein I, has a molecular weight of 32 to 36 kDa and is closely associated in the membrane with LOS. Porin provides channels that allow aqueous solutes to pass through the otherwise hydrophobic outer membrane and is believed to play an important role in pathogenesis. Porin is the product of a gene designated *porB.* Porin proteins occur in two major antigenic classes, designated PorB1A and PorB1B, each of which is composed of many distinct genetic variants. Variations in Por sequence or antigenic types form the basis for the most commonly used gonococcal typing systems.[10] Strains expressing PorB1A and occasionally PorB1B are associated with genotypic resistance of *N. gonorrhoeae* to the bactericidal effect of normal (nonimmune) human serum and, perhaps as a direct result, with an enhanced propensity to cause bacteremia. Porin-related serum resistance is due to binding to loops on the porin protein of the complement downregulatory components C4bp or factor H (fH).[11,12] PorB1A also appears to directly promote invasion of epithelial cells, which also helps explain the propensity for bacteremic dissemination.

Opa proteins are outer membrane proteins with molecular weights of 20 to 28 kDa. They are members of a family of proteins, each produced from its own *opa* gene. The amino-acid sequence of the Opa proteins varies somewhat, primarily because of differences in two hypervariable regions in each protein.[13] Expression of Opa varies because of high-frequency variations in *opa* DNA that result in translational frame shifting. An individual strain of *N. gonorrhoeae* can express none or up to 11 Opa variants but usually not more than 3 at a time. Gonococci isolated from mucosal sites usually express Opa and their colonies are opaque, but most isolates obtained from the cervix during menstruation and isolates from normally sterile sites, such as fallopian tubes, blood, and synovial fluid, generally lack Opa and form translucent colonies. Many Opa proteins increase adherence between gonococci and to a variety of eukaryotic cells, including phagocytes.[14] Certain Opa variants appear to promote invasion of epithelial cells. Two classes of Opa receptor on eukaryotic cells have been identified: heparin-related compounds and CD66, or carcinoembryonic antigen–related cell adhesion molecules (CEACAMs).[15,16] Certain Opa proteins are able to bind to CEACAM receptors on B and T cells, resulting in downregulation of immune responses.[17] This may help account for the poor immune response to natural infection.

Reduction-modifiable protein (Rmp) has a molecular weight of 30 to 31 kDa; is present in all gonococci in close association with porin and LOS; and shows little, if any, interstrain antigenic variation. Rmp can stimulate blocking antibodies that reduce serum bactericidal activity against *N. gonorrhoeae,* which may potentiate infection after sexual exposure to an infected partner.[18] Several other outer membrane proteins have been identified, including multiple iron-repressible proteins, some of which are shared with *N. meningitidis.* Two of the iron-repressible proteins (85 and 110 kDa) constitute a specific receptor for human transferrin,[19] and two others form a receptor for human lactoferrin.[20] The transferrin receptor is required for successful experimental urethral infection, but the role of the lactoferrin receptor is unclear; it does not influence infectivity. Two additional proteins constitute a receptor for hemoglobin.[21] Other proteins are expressed only during anaerobic growth.[22] The ability of *N. gonorrhoeae* to grow anaerobically after removing available oxygen from the microenvironment contributes to its ability to survive in the cervical and vaginal microaerobic environment.[23] Immunoglobulin A1 (IgA1) proteases, present in *N. gonorrhoeae* and *N. meningitidis* but not in nonpathogenic *Neisseria,* are assumed to protect the organism from secretory IgA antibody at mucosal surfaces, but this role has not been proven.

Gonococcal LOS is composed of lipid A and a core oligosaccharide that, in contrast to the polysaccharide of most gram-negative bacteria, lacks O-antigen side chains. Sialylation of LOS core sugars in vitro or in vivo masks epitopes on both LOS and porin and contributes to resistance to bactericidal antibodies.[24] LOS possesses endotoxic activity and contributes to ciliary loss and the death of mucosal cells in the fallopian tube explant model (see Fig. 212.3). LOS core sugars undergo high-frequency phase and antigenic variation in vitro and in vivo, which may contribute to the pathogenesis of infection, including resistance to bacterial anti-LOS antibodies present in normal serum and invasion of epithelial cells.

The peptidoglycan layer of *N. gonorrhoeae* may also contribute to the inflammatory response. The gonococcus has lytic transglycosylases, which produce and release highly inflammatory peptidoglycan monomers.[25] These peptidoglycan fragments are toxic in the fallopian tube explant system and cause complement consumption in vitro. In addition, peptidoglycan fragments have been found in the apparently sterile synovial fluid of patients with gonococcal arthritis-dermatitis syndrome.[26] Gonococci produce a surface polyphosphate that may have capsule-like functions, such as creating a hydrophilic, negatively charged cell surface. However, a carbohydrate capsule analogous to that of *N. meningitidis* or *Streptococcus pneumoniae* is not produced.

Strain Typing

Studies of the clinical manifestations and epidemiology of gonorrhea have been greatly enhanced by the development of reproducible methods for typing *N. gonorrhoeae.* These methods are not available in clinical laboratories, however. They consist of characterization of gonococcal strains based on two primary methods: auxotyping and serotyping using monoclonal antibodies to variable epitopes on the porin protein. In the future, with the introduction of rapid and inexpensive whole-genome sequencing, strains will be compared through use of these methods.

Genetics

Plasmids

Many gonococci possess a 24.5-mDa conjugative plasmid and can thereby conjugally transfer other non–self-transferable plasmids with high efficiency; chromosomal genes are not mobilized. Many gonococci carry a plasmid (Pc^r) that specifies production of a TEM-1 type of β-lactamase (penicillinase). The two most common Pc^r plasmids have molecular weights of 3.2 and 4.4 mDa and are closely related to each other and to similar plasmids found in certain *Haemophilus* spp., including *Haemophilus ducreyi.* In fact, it is suspected that gonococci first acquired Pc^r plasmids from *H. ducreyi.*[27] Pc^r plasmids are commonly mobilized to other gonococci by the conjugative plasmid.

Gonococci with plasmid-mediated high-level resistance to tetracycline, with minimum inhibitory concentrations (MICs) of 16 mg/L or greater, carry the 24.5-mDa conjugative plasmid into which the *tetM* transposon has been inserted.[28] The *tetM* determinant also confers tetracycline resistance to a variety of other bacteria, including some *Streptococcus* and *Mycoplasma* spp. and various genital organisms such as *Gardnerella vaginalis* and *Ureaplasma urealyticum.* Because of its location on the conjugative plasmid, high-level tetracycline resistance is readily transferred among gonococci. The *tetM* determinant functions by encoding a protein that protects ribosomes from the effect of tetracycline. Finally, all gonococci contain a small (2.6 mDa) cryptic plasmid of unknown function.

Chromosomal Mutations and Transformation

Mutations in biosynthetic pathways are common, presumably reflecting the ready availability in vivo of essential nutrients such as amino acids, purines, and pyrimidines at infected mucosal sites. Nevertheless, *N. gonorrhoeae* is not highly mutable in that it lacks error-prone repair systems and is relatively resistant to external mutagenic stimuli such as ultraviolet light. Instead, gonococci have evolved efficient systems

for phase and antigenic variation of surface components (pili, Opa, and LOS) that do not depend on such mutagenic pathways.

Gonococci also use transfer of naked DNA between cells (transformation) to promote genetic variability. The piliated variants of virtually all clinical isolates of *N. gonorrhoeae* are highly competent in transformation, but loss of the ability to express pili is always accompanied by a dramatic reduction in transformation competence. Uptake of transforming DNA is limited to homologous (i.e., gonococcal) DNA, which reflects recognition of a unique nucleotide sequence by a surface receptor.[29] No bacteriophages have been found in *N. gonorrhoeae.*

Chromosomal resistance of *N. gonorrhoeae* to β-lactam antibiotics and the tetracyclines results from interactions among a series of individual mutations, some of which (e.g., the *mtr* determinant) alter the net accumulation of antimicrobial agents inside the cell. The *mtr* locus has been shown to be an efflux pump similar to other membrane transporters.[30] The *penA* locus alters penicillin-binding protein 2 to reduce its affinity for penicillin.[31] For epidemiologic purposes, chromosomal resistance is defined when the MIC is such that clinical failures are common with the maximum practical therapeutic dose, which corresponds to MICs of 2 mg/L or greater for both tetracycline and penicillin G. Clinically significant resistance to the fluoroquinolones, indicated by MICs of ciprofloxacin of 1 mg/L or higher (up to 16 mg/L), result from the additive effects of multiple chromosomal mutations involving the genes *gyrA* and *gyrB,* which code for DNA gyrases, and *parC* and *parE,* which code for topoisomerases.[32]

PATHOBIOLOGY OF GONOCOCCAL INFECTION

An unusual characteristic of gonococcal infection is that the pathobiologic events that result in infection in men and women are different, leading to different clinical expression of the infection.[33]

Infection of the Male Urethra

N. gonorrhoeae infection in men occurs as an acute urethritis, which develops from the concomitant inflammatory response directed at infecting gonococci. One hallmark of gonococcal disease in men is the frequent presence of a purulent urethral discharge, which is associated with polymorphonuclear leukocyte (PMN) influx and shedding of urethral epithelial cells. Human volunteer studies indicate that there is an incubation period from the time of infection to the onset of clinical symptoms of disease; during this time, gonococci cannot be cultured from the urethra for up to 40 hours after the initiation of infection.[34] In vitro infection assays and microscopic analysis of patient exudates indicate that gonococci can enter urethral epithelial cells and are released, and then infected epithelial cells are shed from the mucosal surface to the urethral lumen.[35] Experimental infection in men has also demonstrated that high concentrations of the chemokine interleukin (IL)-8 and cytokines IL-6 and tumor necrosis factor-α (TNF-α) are present within the urethral lumen with progressive gonococcal infection.[36] Recent evidence demonstrates that LOS elicits TNF-α, IL-1β, IL-6, and IL-8 secretion from urethral epithelial cells.[37] Release of cytokines and chemokines from the urethral epithelium may therefore initiate the inflammatory response associated with gonococcal urethritis by triggering PMN influx. PMN influx in conjunction with cytokine release from the urethral epithelium subsequently potentiates the clinical symptoms associated with disease. Unless intercepted by effective antimicrobial therapy, this process is cyclic during the course of infection, with extension into the upper male genital tract. The acquired immune response in humans is ineffective in slowing disease progression or preventing reinfection.

Microscopic examination of urethral exudates from men documented to have culture-proven gonorrhea indicates that gonococci are found within PMNs and urethral epithelial cells.[38] The interaction of gonococci with PMNs is dependent on the presence of Opa, but it does not require pili. Opa proteins can be divided into two broad classes represented by Opa_{50} (i.e., proteins that recognize host cell heparin sulfate proteoglycans [HSPGs]) and Opa_{52} (i.e., proteins that recognize CEACAMs[39]). Experimental infection of men with Opa^- gonococci results in a shift to an Opa^+ phenotype.[36] An Opa^+ phenotype is also prevalent in clinical isolates obtained from men with naturally acquired gonococcal infection. Primary male urethral epithelial cells do not express CEACAMs, and an *N. gonorrhoeae* strain FA1090Δ Opa mutant is not impaired in its ability to cause infection in a human experimental model. This would suggest that the role of Opa proteins in gonococcal urethritis in men resides in their ability to facilitate a gonococcal-PMN interaction. (Accordingly, generating such an inflammatory response may give a survival advantage to the gonococcus because it can replicate within the PMN.[40])

CEACAMs can serve as coreceptors for other cell surface receptors (e. g., integrins) present on professional phagocytic cells. Engagement of CEACAMs sends a priming signal within PMNs that activates adhesion receptors without triggering a respiratory burst or the release of inflammatory mediators. An Opa-CEACAM interaction may therefore enhance gonococcal survival within these cells.

Porin also contributes to the intracellular survival of gonococci within PMNs. Gonococcal porin is unique among gram-negative bacterial porins in its ability to translocate to and insert into a targeted host cell membrane. Within a host cell membrane, porin forms an anion-selective, voltage-gated channel that is modulated through its interaction with adenosine triphosphate or guanosine triphosphate.[41] Insertion of porin into the PMN membrane inhibits degranulation by causing a change in membrane potential without triggering the respiratory burst within these cells.[42] Further modulation of host cell function attributed to porin involves its ability to inhibit phagosome maturation[43] and to downregulate cell surface receptors important to immune function (e.g., FcγRII, FcγRIII, and complement receptors 1 [CR1] and 3 [CR3]).[44] A single primary receptor has not been identified that uniquely modulates the interaction of the gonococcus with PMNs; however, this association occurs independently of CR3.[45] Fcγ-, CEACAMs, HSPGs, and integrin receptors present on the PMN cell surface may all play a role in gonococcal adherence and/or its internalization.

The initial site of gonococcal disease in men is the urethral epithelial cell (Fig. 212.4A). Disease occurs as a sequential process in which an initial interaction occurs between gonococcal pilus and the urethral epithelium. Pilus expression is required for efficient infection of the urethral epithelium. An intimate association between the urethral epithelium and the gonococcus (see Fig. 212.4B) is achieved through the interaction of the asialoglycoprotein receptor (ASGP-R) and gonococcal LOS,[46] a major constituent of the gonococcus cell membrane. Engagement of the ASGP-R by the gonococcus results in pedestal formation beneath the bacterium. Endocytosis ensues primarily because of actin-[47] and clathrin-dependent[35] processes. Endocytosis mediated by the ASGP-R results in endosomal fusion and acidification, which results in clathrin-coat disassembly and uncoupling of the ASGP-R–ligand complex. After gonococcal internalization, ASGP-R is recycled to the urethral cell surface, where it is available to bind more gonococci. Small proportions of infecting gonococci enter urethral epithelial cells by a macropinocytic mechanism, although membrane ruffling is not observed.[48] The intracellular fate of the gonococcus is unclear. The gonococcus does induce antiapoptotic events that prolong the life of the epithelial cell.[49,50]

Adherence of the gonococcus to the ASGP-R is dependent on the presence of an exposed galactose on the terminal lacto-*N*-neotetraose (LNnT) moiety on LOS. This moiety mimics human paragloboside and provides one means by which the gonococcus escapes immune recognition. In addition, the LNnT epitope can serve as a sialic acid acceptor. The presence of sialic acid on gonococcal LOS confers (unstable) resistance to the bactericidal action of normal human serum (i.e., serum resistance). The importance of the LNnT moiety to gonococcal pathogenesis in men can be inferred from human experimental infection studies and from clinical data obtained from men with naturally acquired gonorrhea, which demonstrate that LNnT is selected for in vivo.[51,52] Gonococci bearing the LNnT moiety on their LOS exhibit enhanced infectivity in human volunteer studies.[53] It is thought that serum resistance conferred by LOS sialylation allows a greater proportion of gonococci to survive the innate immune factors in the urethral lumen during disease. Consequently, a lower infectious dose is required to establish disease because a greater proportion of the infection inoculum survives and proliferates.

Sialylation of gonococcal LOS occurs when it is intracellular. LOS sialylation is mediated by gonococcus-encoded sialyltransferase[54–59]

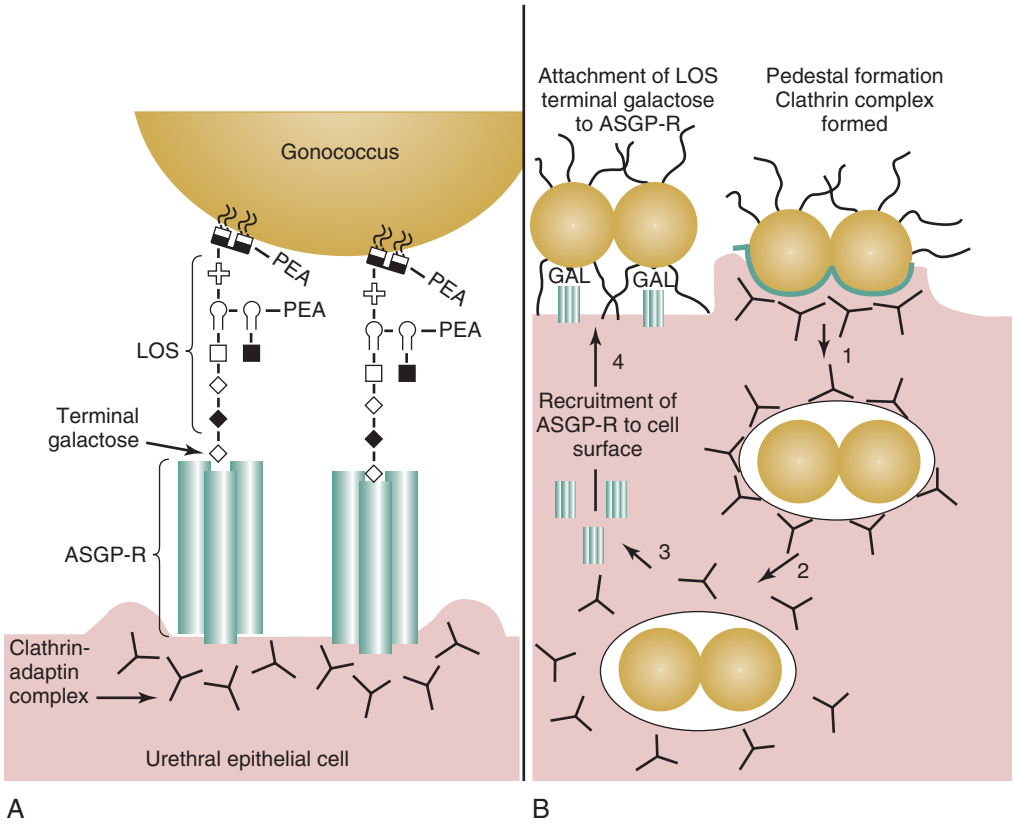


FIG. 212.4 Infection of the urethra by the gonococcus is accomplished by receptor-mediated phagocytosis of the organism by the urethral epithelial cell. (A) Interaction between the asialoglycoprotein receptor *(ASGP-R)* and the terminal galactose of the lipooligosaccharide *(LOS)* initiates a signaling cascade, which results in recruitment of clathrin to the site of ASGP-R, and gonococcus–ASGP-R complexes are internalized in clathrin-coated pits in an actin-dependent process. (B) The bacterium is engulfed by and in close interaction with the epithelial cell membrane and is taken into the cell. The enzyme dynamin cleaves the now-engulfed bacterium from the cell plasma membrane. The bacterium is within a tightly formed endosome, and within this endosome a drop in pH is proposed to release the ASGP-R from the gonococcus surface and clathrin molecules also are released. The receptor clathrin complexes begin to disperse from the bacteria-containing endosome and are available for recruitment back to the cell membrane. This results in further uptake of bacteria at the epithelial cell surface. Sialylated gonococci are eventually released from the urethral epithelium, where they can then be transmitted to a female partner. *GAL,* Galactose; *PEA,* phosphoethanolamine.

present in the gonococcal outer membrane. The gonococcus lacks the ability to synthesize cytidine 5′-monophosphate *N*-acetylneuraminic acid (CMP-NANA) and must parasitize this substrate from its human host.[60,61] Sialylation of the LNnT epitope impairs the ability of gonococci to invade primary urethral epithelial cells and epithelial cell lines, to cause disease in human volunteers, and to be phagocytized by neutrophils. Gonococcal infectivity is restored with sialic acid removal by neuraminidase or by the replication of gonococci within the lumen of the urethra in the absence of host-derived CMP-NANA. Within the lower female genital tract, sialylated gonococci may become modified to enhance disease transmission to men—that is, neuraminidases produced by the vaginal microbiota (e.g., *G. vaginalis*[62]) may remove sialic acid from sialylated gonococci. Cervical epithelia also produce neuraminidase[63]; however, the specificity of this enzyme to cleave endogenous or exogenous substrates exhibits cyclic variability. The level of sialic acid found within the microenvironment of the cervix also exhibits cyclic variation. Neuraminidase and the ASGP-R are also present on human sperm.[64] Sialylated gonococci in proximity to sperm cells may become desialylated through the action of neuraminidase present on these cells. Subsequent gonococcal adherence to the ASGP-R on sperm can then, in turn, facilitate disease transmission.[64]

Infection of the Lower Female Genital Tract

The majority of gonococci transmitted from men to their partners have sialylated LOS. However, the presence or absence of sialic acid on LOS does not influence the interaction of the gonococcus with the cervical epithelium.

In contrast to the overt inflammatory response generated with gonococcal infection of the male urethra, 50% to 80% of women with lower genital tract *N. gonorrhoeae* infection are asymptomatic and 70% to 90% of women with disseminated infection lack signs of genital tract involvement. Analysis of cervical secretions obtained from uninfected women and from women infected with the gonococcus reveal that an antibody response is not generated with uncomplicated infection. These findings are consistent with the ability of the gonococcus to evade and to subvert host immune function.[65]

Within the lower female genital tract, the cervical epithelium provides a source of alternative pathway (AP) complement (C′) activity, albeit at a level only comparable to approximately 10% of that observed for human serum. These components are produced by the cervical epithelia. Within minutes of infection of cervical epithelia, C′ protein C3b is deposited on the lipid A portion of gonococcal LOS[66] and is rapidly inactivated to iC3b (Fig. 212.5A).[67] These data are supported by the predominance of iC3b (in comparison to C3b) on the surface of clinically isolated gonococci.[68,69] The affinity of C′ fH for sialylated LOS[12] and for porin of a PI.A isotype[12] may augment C3b inactivation. However, C3b inactivation occurs in a kinetically similar manner on gonococci of either a PI.A or a PI.B isotype and on sialylated gonococci or on gonococci that are not sialylated.[67]

CR3 serves as the primary receptor for *N. gonorrhoeae* adherence to and invasion of the ectocervix and endocervix (see Fig. 212.5A).[67] Binding of gonococcal pilus to the I-domain of CR3 probably allows the gonococcus to overcome the electrostatic repulsion between its own cell surface and that of the cervical cell. The twitching action of the gonococcal pilus with reduction in pilus length may act to juxtapose the gonococcus

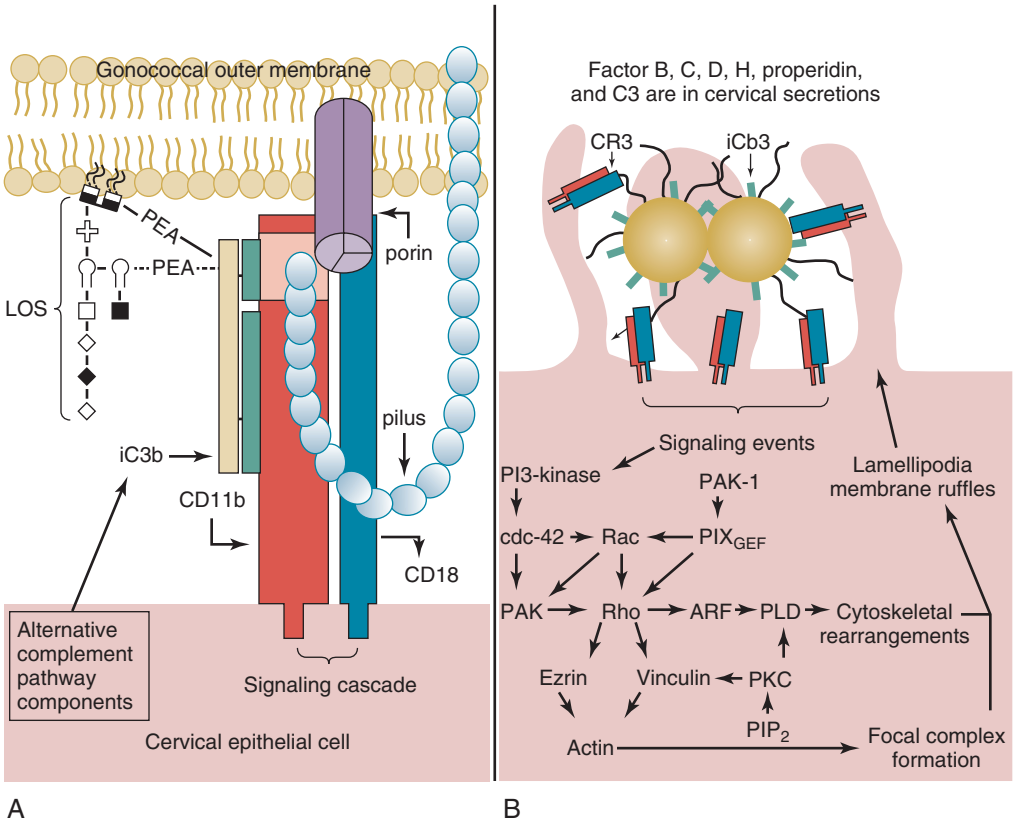


FIG. 212.5 Infection of cervical epithelia by the gonococcus. (A) Alternative complement pathway components are produced and released by the cervical epithelia. On *Neisseria gonorrhoeae* infection, complement is activated. Gonococcal pilus binds to the I-domain (red region on CD11b) of CR3 receptor, which comprises the integrins CD11b and CD14, allowing the bacterium to overcome the electrostatic repulsion between its own cell surface and that of the host cell. As the pilus retracts, this places the gonococcus within proximity to the cervical cell surface where complement components would be sufficient to allow deposition upon the bacterial cell surface. C3b forms a covalent association with amine groups on the deep core region of gonococcal lipooligosaccharide (LOS) and is rapidly inactivated to iC3b. The affinity of gonococcal porin for factor H may augment iC3b formation. The proximity of porin to LOS in the outer membrane may spatially favor the intimate association between iC3b and porin with the CR3 I-domain. (B) Engagement of CR3 triggers a complex signal transduction cascade mediated by the Src-family tyrosine kinases and Rho GTPases. These processes result in vinculin- and ezrin-enriched focal complex formation and membrane ruffling, that is, the trigger mechanism of invasion. Secreted *N. gonorrhoeae* phospholipase D modulates cervical cell signal transduction by playing a role in CR3 recruitment to the cervical cell surface and by modulating cytoskeletal reorganization. Additionally, this protein augments intracellular survival of gonococci after their internalization within macropinosomes. Sialylated gonococci are eventually released from the cervical cell, where they are free to invade more of the cervical epithelia or where they become primed for transmission to the male urethra on sialic acid removal by sialidases present within the female genital tract. However, sialic acid on the gonococcus surface does not influence the ability of these organisms to infect the human cervical epithelial cell.

at the cervical cell surface, where C′ concentrations would be expected to allow efficient opsonization for the subsequent intimate adherence of iC3b on the organism surface and gonococcal porin to the I-domain. Thus, binding of the gonococcus requires the cooperative action of iC3b bound to the gonococcal surface in conjunction with gonococcal porin and pilus.[67] Engagement of CR3 results in a complex signaling cascade in which a vinculin- and ezrin-enriched focal complex formation occurs before membrane ruffle formation.[70] A signal transduction cascade that is dependent on the activation of wortmannin-sensitive kinases (i.e., phosphatidylinositol 3-kinase or mitogen-activated protein kinases)[70] and Rho GTPases[67] initiates ruffling (see Fig. 212.5B). Gonococci are then internalized within macropinosomes.[70]

On infection of cervical epithelia, gonococci release a phospholipase D homologue that gains access to the cervical intracellular environment nonspecifically through macropinocytosis of gonococci.[71] Gonococcal phospholipase D (NgPLD) appears to promote infection of the cervical epithelium in several ways. Data indicate that this secreted gonococcal protein augments signaling events that trigger CR3 mobilization to the cervical cell surface.[71] This ensures gonococcal receptor availability and, consequently, efficient targeting to and association with the cervical cell surface. NgPLD also modulates cervical cell signal transduction events, leading to membrane ruffling. Mutant gonococci that lack functional NgPLD activity do not elicit membrane ruffling, and they are impaired in their ability to associate with and to invade primary human cervical cells.[71]

As with invasion of male urethral epithelial cells, the intracellular fate of gonococci within the cervical epithelium is unclear. Ligand binding to the I-domain of CR3 does not invoke a proinflammatory response in professional phagocytic cells. Consequently, the ability of gonococci to subvert cervical cell signal transduction cascades and the complement system in such a manner to allow a cooperative mechanism of CR3-mediated adherence to and invasion of the cervical epithelium may also enhance its survival within the lower female genital tract. Gonococcal invasion in the absence of a respiratory burst increases the number of gonococci that survive intracellularly, whereas inactivation of the C′ system enhances gonococcal survival extracellularly. Consequently, subversion of host cell signal transduction and the C′ system by the gonococcus within the lower female genital tract allows this bacterium to obtain a carrier-like state. Ascending infection of the uterus and fallopian tubes may occur as a consequence of hormonal changes that alter the mucosal epithelia, molecules available for gonococcal use, and/or virulence factors expressed by the gonococcus. In this regard, menses is associated with an increased risk to women for gonococcal PID and disseminated infection. C3 production by the cervical epithelium exhibits cyclic variability, and the highest levels of C3 are detected during menses. A correlation can also be made between the presence or absence of

Opa and the site of gonococcal isolation. Opa^- (or transparent) gonococci predominate in the fallopian tubes and in the cervix at the time of menses. Conversely, Opa^+ gonococci predominate in the male urethra and the cervix at the time of ovulation.

Infection of the Upper Female Genital Tract

Approximately 45% of women with gonococcal cervical infection will develop an ascending infection,[72] the prerequisite to PID. Ascent to the upper female genital tract may be facilitated by the ability of gonococci to exhibit twitching motility in conjunction with hormonal changes, which influence the expression of C′ and molecules serving as gonococcal receptors within the female genital tract. Human expression of CR3 progressively decreases in an ascending manner from the ectocervix to the fallopian tubes.[67] Conversely, expression of the lutropin receptor (LHr) increases in an ascending manner from the endometrium to the fallopian tubes, and expression is upregulated during menses.[72,73] The LHr serves as a receptor for gonococcal invasion of fallopian tube epithelia.[74] The interaction of the gonococcus with the LHr increases the invasive character of the gonococcus for the fallopian tube epithelia and therefore is said to occur in a contact-inducible manner.[74,75] Human chorionic gonadotropin (hCG), a ligand for LHr, can competitively inhibit the LHr-gonococcus interaction. These data suggest that gonococci possess a surface molecule that mimics hCG. Recently, Spence and coworkers[75] reported that this hCG-like molecule is the ribosomal protein L12. L12 is shown to facilitate gonococcal transcytosis through the fallopian tube epithelia. The LHr is also present on the human uterus, placenta, decidua, and fetal membranes.[76] Hypothetically, a gonococcus-LHr interaction occurring on decidua and placental membranes could result in severe complications of disease and may in part contribute to the increased risk for spontaneous abortion associated with *N. gonorrhoeae* infection.

Gonococcal adherence via the LHr occurs selectively on nonciliated cells[7,74]; however, it is the ciliated cells of the fallopian tube epithelia that are subsequently shed.[77] If the infection is left untreated, complete loss of ciliary action can occur. Cytotoxicity of ciliated cells is attributed to gonococcal peptidoglycan[78] and LOS[79,80] either directly or indirectly through the induction of increased production of the inflammatory cytokine TNF.[81] The loss of ciliated cells within the fallopian tube provides the gonococcus access to subepithelial tissues. Access to subepithelial tissue is also obtained with invasion of nonciliated cells, after which gonococci are transcytosed to the basal lateral surface of these cells and released. In Hec1B cells, the gonococcal protein L12 mediates transcytosis to the basal lateral surface[75]; however, this has yet to be demonstrated in a fallopian tube organ model. Sialylation of intracellular gonococci (before their exocytosis) might prime these organisms for disseminated infection because of the increased serum resistance.

THE HUMAN IMMUNE RESPONSE TO *NEISSERIA GONORRHOEAE*

Adaptive Immune Response

Studies of human immune responses to *N. gonorrhoeae* have yielded little information about the factors necessary for development of a protective response. It has been recognized for years that recurrent infection by the gonococcus after successful treatment is a common event. The diary of Boswell describing his frequent recurrences of infection are testimony to this fact.[82] Some of his infections persisted for months before clearing. Modern studies have shown that the gonococcus can persist in asymptomatic women and men for over 6 months, and probably considerably longer.[1,2] Using a human experimental model of infection, by challenging subjects with the homologous organism[3] after clearing the first infection with antibiotics, investigators showed that recurrent infection can occur. The inability of the human to readily clear the infection and the reports of long-term carriage and reinfection by the same organism all are indicative of very poorly protective immune response to the gonococcus. Studies of the human immune response focused on the evolution of serum antibodies to pilus and porin after infection but yielded little evidence that a protective response could be developed or sustained.[4,5] Despite these observations, a study was undertaken to protect individuals from gonorrhea through use of a pilus-based vaccine. Analysis of the results of this study indicated that the vaccination was completely ineffective.[6] Since the application of molecular techniques to the gonococcus and the elucidation of the genome, a number of organism factors have been found that allow the organism to evade the host immune response. These include the ability of a large number of surface structures to undergo phase and antigenic variation,[7–9] the molecular mimicry by surface glycolipids of human antigens,[10–12] and horizontal gene exchange.[13] In addition, studies in mice suggest that the gonococcus may directly inhibit the adaptive and innate immune responses.[14] The mechanisms for these effects are not known. Analysis of individuals receiving the four-component meningococcal B vaccine suggested that limited protection against gonococcal infection occurred.[15] These results need to be confirmed in future studies.

Innate Immune Response

More recent interest in the innate immune response has stimulated similar studies on *N. gonorrhoeae*. The hallmark of symptomatic gonococcal infection in humans is a robust exudative PMN response. In spite of this, the gonococcus persists and the inflammatory response can continue untreated for prolonged periods.[82] Studies have shown that gonococci can delay fusion with primary granules in the neutrophil[16] and that lytic enzymes involved in peptidoglycan biosynthesis impart resistance to killing of the gonococcus by lysozyme and human neutrophils.[17] The gonococcus appears to be immune to the effects of antimicrobial peptides as a result of a variety of mechanisms from novel pumps to modifications of lipid A structures by phosphoethanolamine.[18–22] Studies have shown that in vivo the sialylated LOS enhances deposition of fH, an inhibitor of the alternative pathway of complement to the bacterial surface. This results in the degradation of C3 to inactive C3b and loss of complement lytic activity.[23–25] Taken together, the findings of previous studies of the immune response, both adaptive and innate, reveal an organism that is remarkably adapted to the human host.

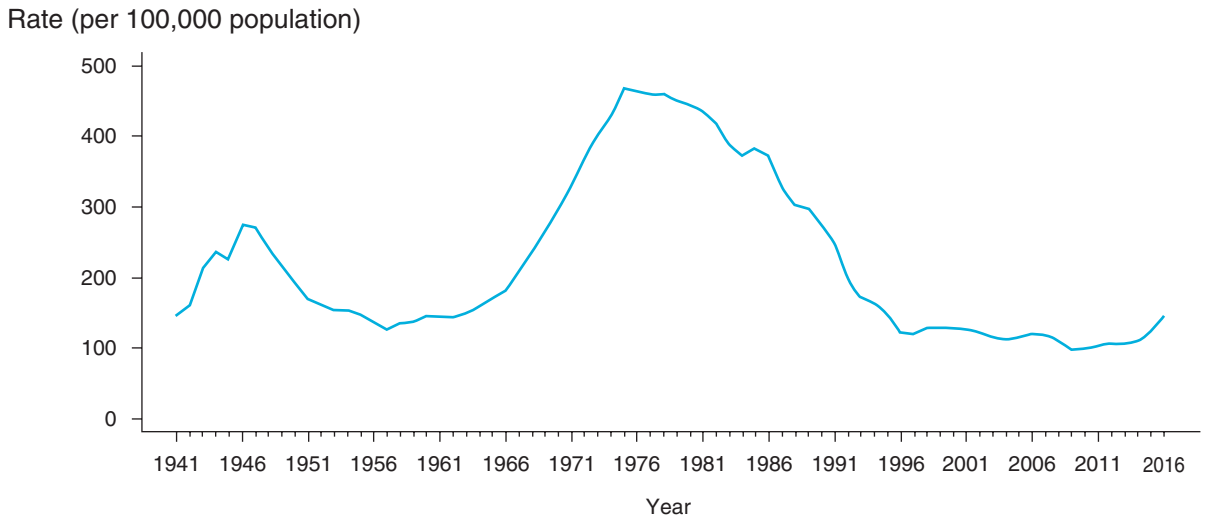
EPIDEMIOLOGY

Incidence

Many industrialized countries but few developing ones possess reporting systems that permit reliable estimates of the incidence of gonorrhea. The number of reported cases in the United States increased from approximately 250,000 cases in the early 1960s to a high of 1.01 million cases in 1978. The peak incidence of reported infection in modern times, 468 cases per 100,000 population, occurred in 1975 (Fig. 212.6). The incidence then declined rapidly, largely the result of systematic public health prevention measures implemented in the 1970s. The decline resulted in a historical low of 98.1 cases per 100,000 in 2009. After slight increases each subsequent year, rates increased markedly during 2015 and 2016: the overall rate increased by 18.5% (22% among men and 13.8% among women) to a total of 468,514 cases for an overall rate of 145.8 cases per 100,000.[83] These statistics likely represent a significant underestimate of the true burden of disease.

The incidence of gonorrhea is substantially lower in all countries of western Europe than in the United States, but high and rising rates have been documented in eastern Europe. In the past, the highest incidences of gonorrhea and its complications occurred in developing countries, and this likely remains true in some areas of the world, with particularly devastating consequences for women and their reproductive health. However, extensive use of antibiotic regimens for syndromic management of genital complaints, including urethral and vaginal discharge and PID, has effected a decline in gonorrhea prevalence in many countries.[84]

In the United States, the highest rates occur in 15- to 24-year-old females and males, but after adjustment for sexual experience, the highest rates are seen in sexually active 15- to 19-year-old females (Fig. 212.7).[85] According to the population-based National Health and Nutrition Examination Survey, from 1999 to 2008 the prevalence remained higher among non-Hispanic blacks relative to whites.[86] The Add Health study of young adults showed similar results. Among 12,548 adults aged 18 to 26 years, the prevalence of gonorrhea was 0.43% (95% confidence interval [CI], 0.29%–0.63%), and it was strikingly higher in blacks than in whites (2.13%; 95% CI, 1.46%–3.10%).[87]



Note: Data collection for gonorrhea began in 1941; however, gonorrhea became nationally notifiable in 1944. Refer to National Notifiable Disease Surveillance System (NNDSS) website for more information: https://wwwn.cdc.gov/nndss/conditions/gonorrhea/

FIG. 212.6 Incidence of reported gonorrhea per 100,000 residents, United States, 1941 to 2016. *(From Centers for Disease Control and Prevention. Sexually Transmitted Disease Surveillance, 2016. Atlanta: US Department of Health and Human Services; 2017.)*

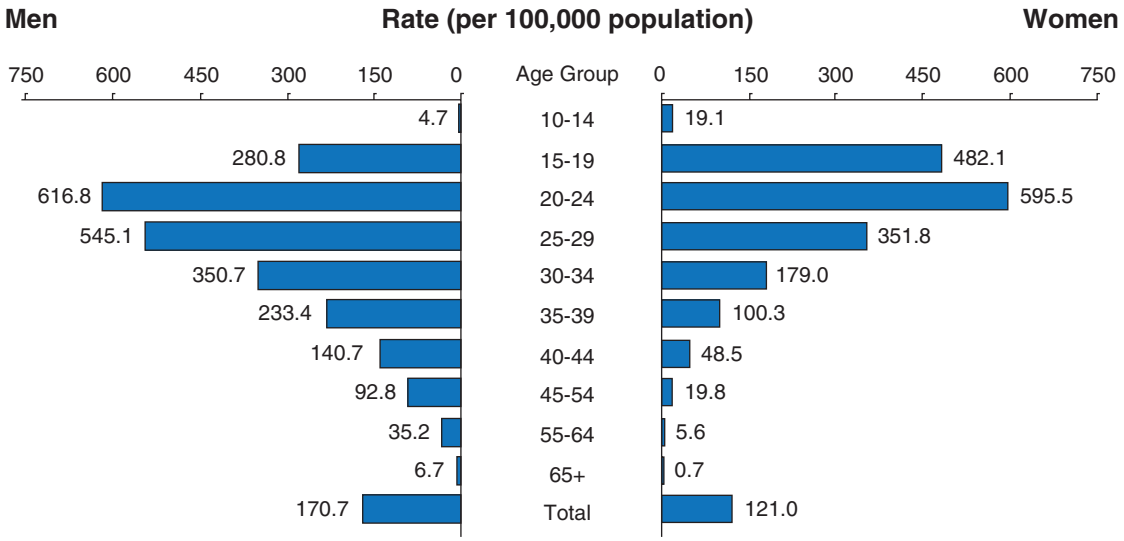
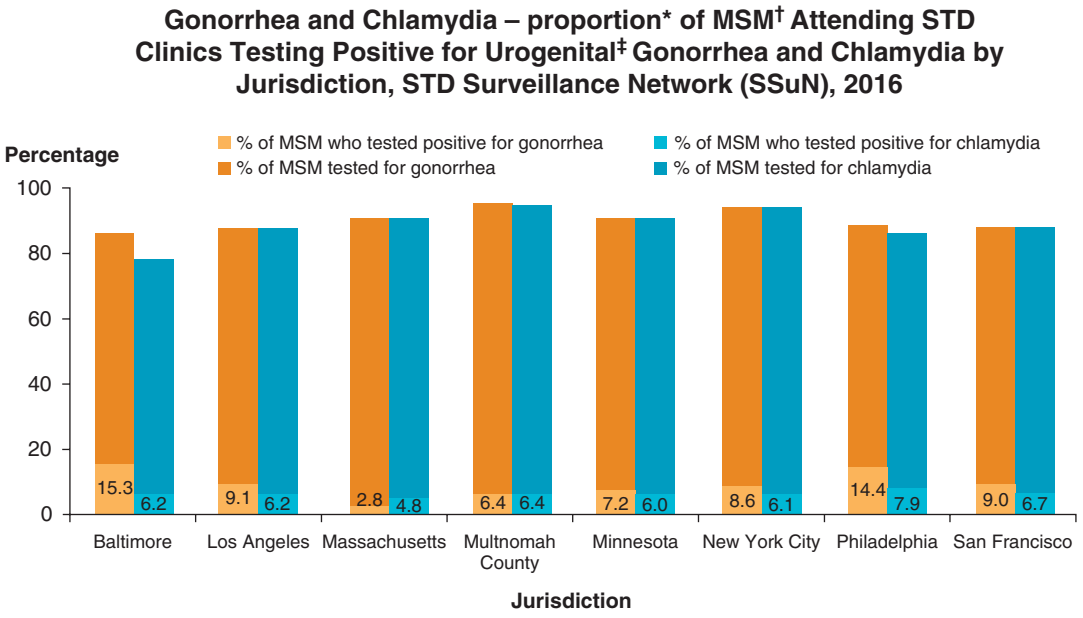


FIG. 212.7 Distribution of reported gonorrhea incidence according to age and sex, United States, 2016. *(From Centers for Disease Control and Prevention. Sexually Transmitted Disease Surveillance, 2016. Atlanta: US Department of Health and Human Services; 2017.)*

Overall, more cases of gonorrhea are reported in men than in women, likely reflecting both a greater ease of diagnosis in men and a substantially higher rate of infection in men who have sex with men (MSM) than in heterosexual men and women. The incidence of gonorrhea among MSM in industrialized countries has been relentlessly increasing in the past several years, presumably a result of behavioral disinhibition in response to improved therapy and survival of people with human immunodeficiency virus (HIV) infection and, more recently, the increasing use of HIV preexposure prophylaxis.[88,89] These trends are evident in data from the Centers for Disease Control and Prevention (CDC) STD Surveillance Network (SSuN), a representative sample of gonorrhea case reports from across the United States. In 2016 the proportion of MSM with rectal or urethral gonorrhea and chlamydia at sexually transmitted disease (STD) clinics varied by site (Figs. 212.8). Across all SSuN sites in 2016, 44.7% of gonorrhea cases were estimated to be among MSM or men who reported having sex with both men and women. Among six sites that continuously participated from 2010 through 2015, estimated rates among MSM increased from 1369 cases per 100,000 to 3435 cases per 100,000 in that timeframe, suggesting that population-based estimates align with those from clinic-based sites.[90]

The rate of gonorrhea in African-American populations in the United States is almost 25 times higher than that in whites or people of Asian ancestry; Hispanics and Native Americans experience intermediate rates (Table 212.1). Only a small portion of these differences can be explained by greater attendance of nonwhite populations at public clinics, where case reporting is more complete than in private health facilities. Race and ethnicity are demographic markers of increased risk, not factors that directly denote a high risk for gonorrhea or other STDs. Other markers of gonorrhea risk in the United States include lower socioeconomic attainment, lesser education, residence in the southeastern part of the country, unmarried status, and illicit drug use. Contrary to popular perceptions, the population-based incidence of gonorrhea is as high in many rural settings in the United States as in urban ones. Differing incidence rates among population subgroups are related less to variations in numbers of sex partners than to complex and poorly understood differences in sex partner networks, access to health care, and related

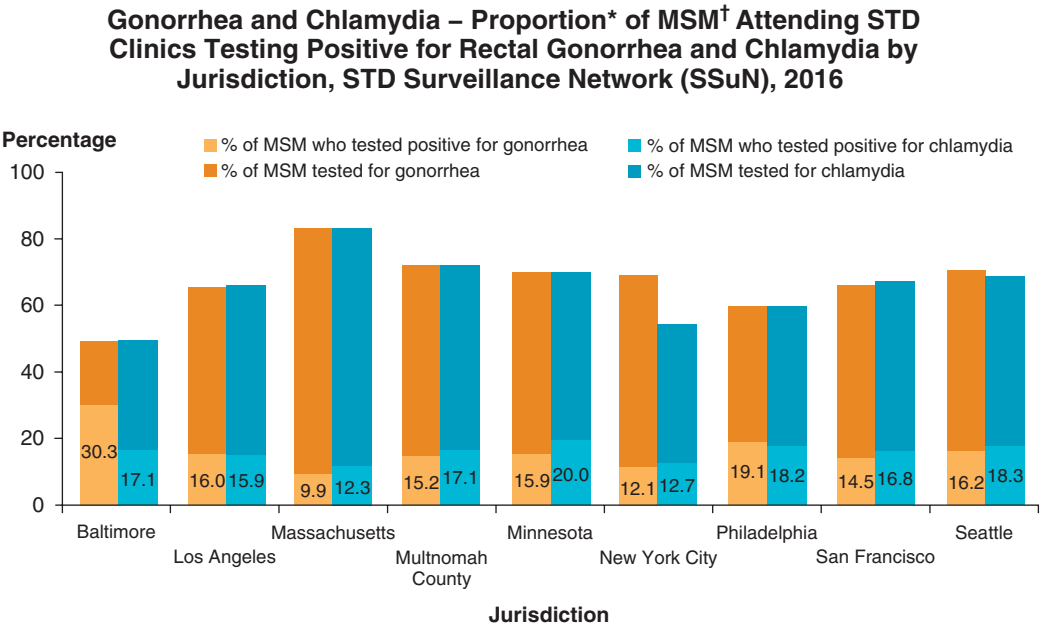


* Results based on data obtained from 22,404 patients tested for urogenital gonorrhea and 22,152 patients tested for urogenital chlamydia attending SSuN STD clinics in 2016; data from Florida and Seattle were not available.

† MSM = Gay bisexual, and other men who have sex with men (collectively referred to as MSM).

‡ Urogenital includes results from both urethral and urine specimens.

A



* Results based on data obtained from 17,467 tested patients tested for rectal gonorrhea and 17,445 patients tested for rectal chlamydia attending SSuN STD clinics in 2016; data from Florida were not available.

† MSM = Gay, bisexual, and other men who have sex with men (collectively referred to as MSM).

B

FIG. 212.8 (A) Proportion of men who have sex with men *(MSM)* with urethral gonorrhea or chlamydia in the Sexually Transmitted Disease Surveillance Network *(SSuN),* 2016. (B) Proportion of MSM with rectal gonorrhea or chlamydia in the SSuN, 2016. *(From Centers for Disease Control and Prevention. Sexually Transmitted Disease Surveillance, 2016. Atlanta: US Department of Health and Human Services; 2017.)*

societal factors. For example, a detailed analysis of increasing gonorrhea incidence in California from 2003 to 2005 raised the importance of contact with a recently incarcerated partner as a major risk and highlighted the relatively understudied contribution of this infection in correctional facilities settings.[91] Moreover, in a large prospective study of 4843 women tested annually for gonorrhea with incidence of 3.5% person-years at risk, number of reported recent sex partners was not independently significantly associated with acquisition.[92] In an elegant analysis, Gomez-Gardenes and colleagues demonstrated that failure to account for these differences in the structure of male-male, female-male, and male-female transmission models can have marked consequences for estimation of transmission dynamics.[93] The demographic predictors of gonorrhea throughout the world are qualitatively similar to those in the United States.

Transmission

The main risk factor for acquiring gonorrhea is sexual intercourse with an infected partner. The risk for transmission of *N. gonorrhoeae* from an infected woman to the urethra of her male partner is approximately 20% per episode of unprotected vaginal intercourse and increases to

TABLE 212.1 Reported Cases and Rates of Gonorrhea According to Race and Ethnicity, United States, 2016

RACIAL OR ETHNIC GROUP	REPORTED CASES	CASES PER 100,000 POPULATION	RATE RATIO
White, non-Hispanic	110,311	55.7	Referent
Black, non-Hispanic	192,114	481.2	8.6
Hispanic	54,299	95.9	1.7
American Indians/Alaska Natives	5757	242.9	4.3
Asians	4934	28.3	0.5
Native Hawaiians/Other Pacific Islanders	928	165.8	3.0
TOTAL	468,514	145.8	

From Centers for Disease Control and Prevention. Sexually Transmitted Disease Surveillance, 2016. Atlanta: US Department of Health and Human Services, 2017.

60% to 80% after four or more exposures.[94] The risk for male-to-female transmission has been less well studied but probably approximates 50% to 70% per contact.[95] Transmission by anal intercourse is efficient, but the risk per episode has not been quantified. Transmission occurs less readily by fellatio, especially from the oropharynx to the urethra, and transmission in either direction by cunnilingus is believed to be rare. Whether women using hormonal contraception are at increased risk for gonorrhea is not known; if so, it is likely that the magnitude of the effect is small.[96]

The incubation period is brief, and prominent genital symptoms often promptly bring infected people to treatment so that the mean duration of gonorrhea is short, typically several days in men and often less than 2 weeks in women. The approximate 50% transmission efficiency of uncomplicated gonorrhea through heterosexual intercourse dictates that especially high rates of sex partner change—an average of two new partners within the 1- or 2-week interval between acquisition of infection and its resolution, equivalent to 50 or more partners per year—are required to sustain transmission in a population. People who have unprotected intercourse with new partners with sufficient frequency to maintain a stable prevalence in the community are defined as core transmitters. Originally developed as a mathematical model, the core transmission hypothesis has been empirically confirmed by several studies,[97] and a central focus of gonorrhea control is to identify the core group and to target members for case finding, treatment, and other prevention strategies. Demographic and social characteristics that directly or indirectly influence the frequency with which new partners are acquired include young age, low educational and socioeconomic levels, commercial sex, illicit drug use, and similar factors. Other characteristics of core transmitters include poorly understood psychosocial determinants of partner selection, cultural factors that affect the response to symptoms, and reduced access to health care (whether real or perceived). A recent variation on the core group concept is the "risk space" hypothesis—namely, that the locations where risk populations live and the sites where sexual exposures occur may be equally or more important determinants of STD incidence than other demographic predictors. Analysis of sexual networks in clusters of gonorrhea is increasingly important in understanding these processes and indicates, for example, that the likelihood that an urban African-American adolescent residing in a high–STD prevalence community will be exposed to an STD is related to the presence of sexual links between his or her recent sex partners and the community.[98,99]

Gonorrhea and other STDs are usually transmitted by people with asymptomatic infections or by those who have symptoms that they ignore or discount.[100] Nevertheless, most people with new genital symptoms cease sexual activity and seek care. It follows that many transmitters belong to a subset of infected people who lack or ignore symptoms. This concept underlies the importance of taking active steps to ensure treatment of the sex partners of infected people, who often will not spontaneously seek health care.

Antimicrobial Resistance

As for most bacterial pathogens, the antimicrobial susceptibility of *N. gonorrhoeae* has evolved under the influence of antimicrobial therapy, and we are now facing a critical juncture as the emergence of resistance to cephalosporins evolves. Unfortunately, this is a clear model for how resistance emerges in the modern age. Initially, gonococci were almost uniformly susceptible to sulfonamides, penicillin, tetracyclines, macrolides, and fluoroquinolones, but none of these is now suitable for routine therapy for uncomplicated gonorrhea in most of the world. Declining susceptibility to penicillin, now attributed to chromosomal mutations, was documented almost immediately after the drug was introduced in the 1940s, but for almost 3 decades penicillin remained useful despite gradually rising relative resistance that necessitated incrementally higher doses and cotreatment with probenecid to enhance and prolong blood levels.

Two nearly simultaneous developments rendered the penicillins unsuitable for routine gonorrhea therapy worldwide. Most dramatic was the appearance in the 1970s of β-lactamase–producing strains of *N. gonorrhoeae* bearing plasmids with the Pc^r determinant, followed by worldwide dissemination within a decade. Several plasmid variants now carry the Pc^r determinant, including the still dominant 4.4- and 3.2-mDa plasmids originally associated with Asia and Africa, respectively.[101] Second, in the 1970s and 1980s, chromosomal resistance progressed to the point that treatment failures became common with the maximum practical single-dose regimens of procaine penicillin, ampicillin, or amoxicillin.

Strains of *N. gonorrhoeae* with plasmid-mediated high-level tetracycline resistance were first documented in the United States in 1985. The location of the responsible *tetM* gene on the conjugative plasmid probably contributed to especially rapid worldwide spread of such strains. The prevalence of tetracycline-resistant *N. gonorrhoeae* (TRNG) peaked in 1997 at 7.3% and decreased for several years. Fortunately, the tetracyclines are no longer recommended and are little used as sole therapy for gonorrhea.

The mutations responsible for chromosomal resistance include *mtr,* which results in increased efflux of several antibiotics and other toxic compounds, such as fatty acids and bile salts; *penA,* which modifies the affinity of penicillin-binding proteins to β-lactam antibiotics; and *penB,* which alters the ability of antibiotics to transit the cell membrane through the porin protein.[30] Simultaneous mutations are common, with additive effects that result in resistance to several antibiotics and to all or most members of each affected drug class. Thus, simultaneous chromosomal resistance to penicillin, the tetracyclines, and macrolides is common. Infection with chromosomally resistant *N. gonorrhoeae* is especially common in MSM. This likely occurs because rectal infection is required for propagation of gonorrhea in MSM and because fecal bile salts and fatty acids confer selection pressure for *mtr.*[102]

As for penicillin and other drugs, low-level resistance to the fluoroquinolones began to appear in *N. gonorrhoeae* almost immediately after they were introduced and soon was observed sporadically throughout the world. Clinically significant resistance, with ciprofloxacin MICs of 1 to 16 mg/L, began to evolve soon thereafter owing to the additive effects of multiple chromosomal mutations, particularly in the DNA gyrase complex (*gyrA* and *gyrB*) and topoisomerases (*parC* and *parE*).[32] Ciprofloxacin MICs of 4 mg/L or greater are associated with at least 50% rates of treatment failure with the recommended regimens of ciprofloxacin or other fluoroquinolones.[103] In the Philippines, the prevalence of high-level resistance to the fluoroquinolones among people with gonorrhea increased from 12% in 1994 to more than 70% in 1996 and 1997,[104] and similar prevalences are now the norm throughout Asia and the Pacific. The proportion of isolates derived from the US Gonococcal Isolate Surveillance Project (GISP) identified as quinolone resistant was 19.2% in 2014 and is notably higher among MSM (30.0% compared with 12.7% in men who reported sex only with women).[105] Such increases prompted the CDC to recommend in 2007 that fluoroquinolones no longer be used to treat gonorrhea in the United States.

Most recently, isolates with resistance to both azithromycin and, most alarmingly, third-generation cephalosporins have emerged.[106] Starting in 2009, the percentage of GISP isolates with cefixime MICs of 0.125 μg/mL increased each year from 1.4% to 1.7% in 2011, decreased to 0.4% in 2013, and increased again to 0.8% in 2014[105] (Fig. 212.9). As was the case with fluoroquinolones, isolates obtained from MSM demonstrate this trend more dramatically. These trends prompted the CDC in August 2012 to modify its STD treatment guidelines for gonorrhea to recommend treatment with parenteral ceftriaxone and to discourage the use of oral cefixime.[107] Moreover, the evolving resistance to cephalosporins detected through routine monitoring has been accompanied by increasing numbers of reports of frank treatment failure, including an increasing number of cases in which parenteral ceftriaxone failed.[108] Emerging data suggest that outbreaks linked to cephalosporin-resistant gonococci may be clonal in origin, which may offer some slim hope for bringing these trends under control in the future.[109–112] Identified mechanisms of cephalosporin resistance are chromosomally mediated, most commonly in the *penA* region, and affect binding of the drug at the β-lactam pocket. However, other mutations are likely needed to effect significant resistance, and the genetics of this process are under intense study.

Finally, the emergence of cephalosporin resistance has been paralleled by a similar trend for azithromycin, with attendant reports of treatment failure and documentation of resistant isolates[113,114] (Fig. 212.10). In 2013, this trend as measured by GISP was stable, with the proportion

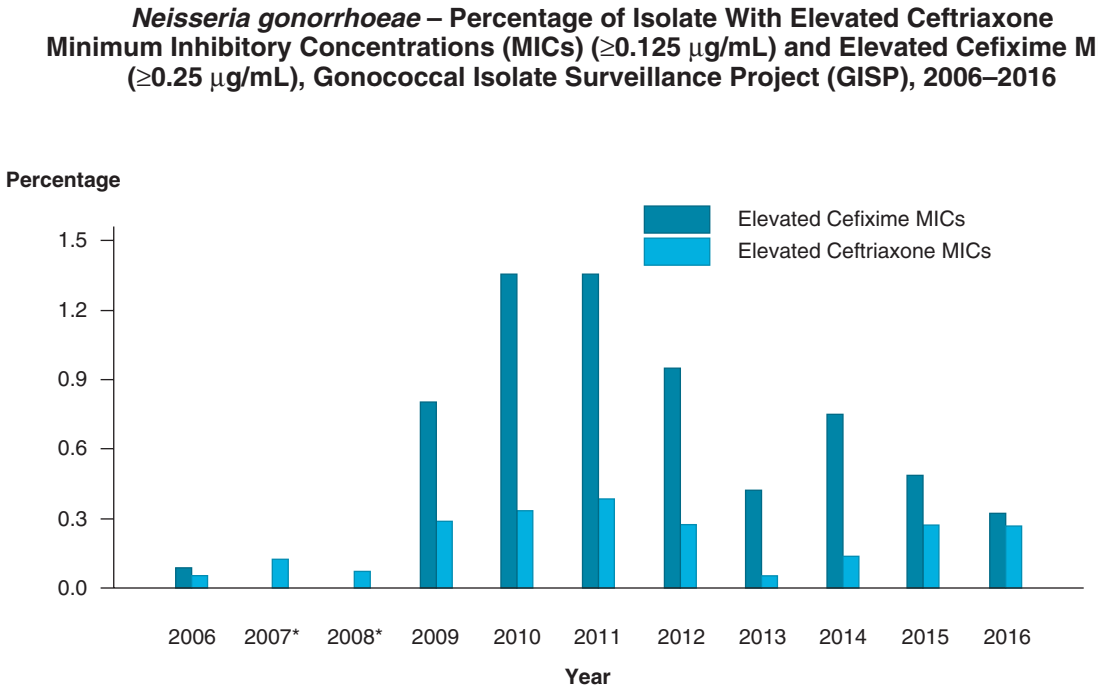


FIG. 212.9 Distribution of minimum inhibitory concentrations *(MICs)* to ceftriaxone in the Gonococcal Isolate Surveillance Project, 2006 to 2016. *(From Centers for Disease Control and Prevention. Sexually Transmitted Disease Surveillance, 2016. Atlanta: US Department of Health and Human Services; 2017.)*

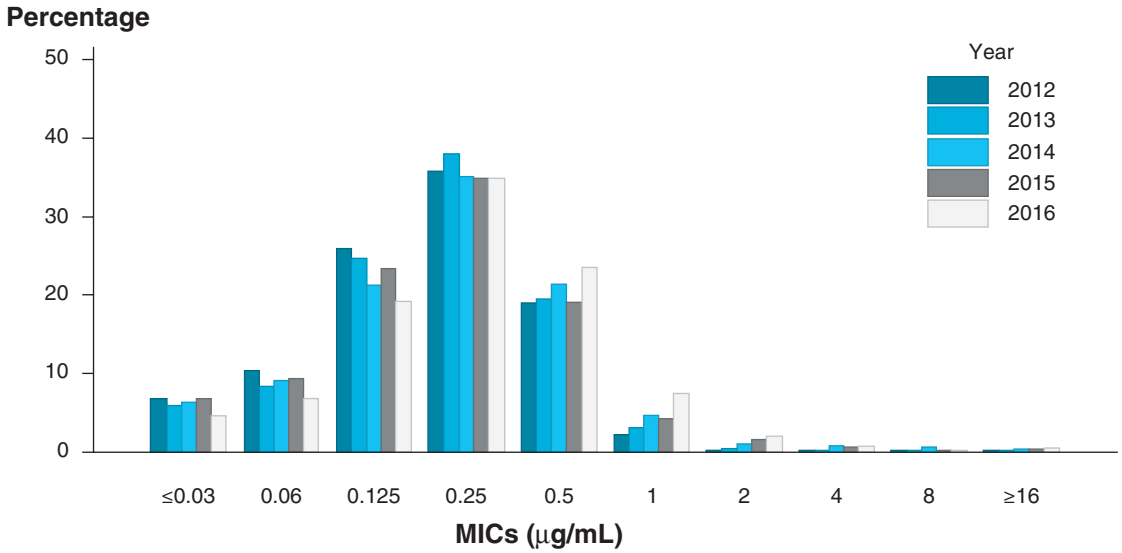


FIG. 212.10 Distribution of minimum inhibitory concentrations *(MICs)* to azithromycin in the Gonococcal Isolate Surveillance Project, 2012 to 2016. *(From Centers for Disease Control and Prevention. Sexually Transmitted Disease Surveillance, 2016. Atlanta: US Department of Health and Human Services; 2017.)*

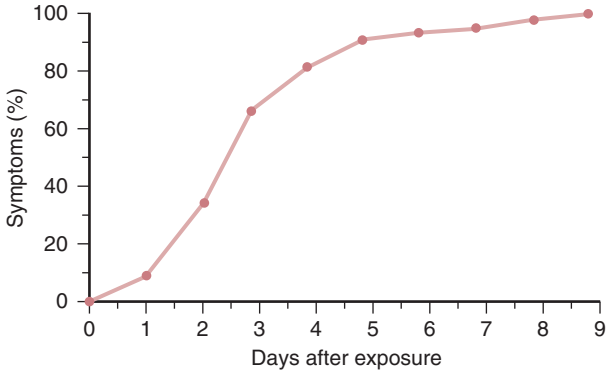


FIG. 212.11 Incubation period in 44 men with gonococcal urethritis. *(From Harrison WO, Hooper RR, Weisner PJ, et al. A trial of minocycline given after exposure to prevent gonorrhea.* N Engl J Med. *1979;300:1074–1078.)*

of isolates with azithromycin MICs greater than 2.0 μg/mL at 0.6%, relatively unchanged from 0.5% in 2010.[115] Unfortunately, this trend has progressed to the point at which some have proposed entirely eliminating this antibiotic as treatment, including in combination with ceftriaxone. In Seattle, Washington, from 2014 to 2016, 5% of gonorrhea cases among MSM had reduced azithromycin susceptibility, defined as MIC ≤1 μg/mL,[116] a trend also reported in Europe.[117]

CLINICAL MANIFESTATIONS

Genital Infection in Men

Uncomplicated Infection

Acute urethritis is the predominant manifestation of gonorrhea in men (see Chapter 109). The incubation period is typically 2 to 5 days but ranges from 1 to 10 days or longer (Fig. 212.11). Urethral discharge and dysuria, usually without urinary frequency or urgency, are the major symptoms. The discharge may initially be scant and mucoid, but within 1 or 2 days it becomes overtly purulent. These observations have been confirmed in studies of experimental gonococcal urethritis in humans.[118] Compared with nongonococcal urethritis, the incubation period of gonorrhea is shorter, dysuria is usually more prominent, and the discharge is generally more profuse and purulent (Fig. 212.12). However, exceptions are common, and a small proportion of men with urethral gonorrhea remain asymptomatic and lack signs of urethritis.[100] The symptoms of urethral gonorrhea depend in part on the infecting organism. Some Por IA serovars and the AHU$^-$ and related auxotypes of *N. gonorrhoeae* are more frequently associated with asymptomatic urethral infection in men than are other gonococcal types.[119] The decline in the prevalence of such strains in North America probably has been accompanied by a decreasing frequency of asymptomatic gonorrhea in men. Most cases of untreated gonococcal urethritis resolve spontaneously over several weeks.

Localized Complications

Acute epididymitis is the most common complication of urethral gonorrhea but now is uncommon in industrialized countries; most cases of epididymitis in young men are due to *Chlamydia trachomatis* (see Chapter 112).[120] Penile edema without other overt inflammatory signs ("bull-headed clap") is occasionally seen in gonococcal or nongonococcal urethritis. Penile lymphangitis, periurethral abscess, acute prostatitis, seminal vesiculitis, and infections of Tyson and Cowper glands are uncommon complications. Urethral stricture as a result of gonorrhea is also uncommon. Although once considered a common consequence of gonococcal urethritis, many strictures in the preantibiotic era might have resulted primarily from treatment with urethral irrigation with caustic solutions, such as silver nitrate or potassium permanganate, rather than from gonorrhea.

Uncomplicated Urogenital Infection in Women

The primary loci of genital infection in women are the columnar epithelial cells that line the endocervix (see Chapter 110). *N. gonorrhoeae* can often

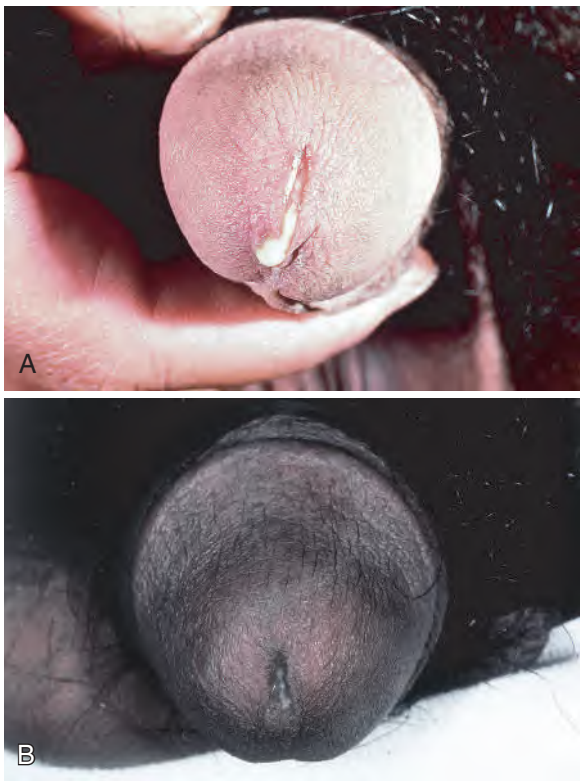


FIG. 212.12 Gonococcal urethritis. (A) Purulent exudates due to gonorrhea. (B) Mucopurulent discharge mimicking the usual appearance of nongonococcal urethritis due to *Chlamydia trachomatis* and other pathogens.

be recovered from the urethra or rectum and occasionally from the periurethral (Skene) glands and the ducts of Bartholin glands, but these are rarely the sole infected sites except in women who have undergone hysterectomy. The vagina per se is not infected in sexually mature women because under the influence of estrogen the squamous epithelium of the vaginal mucosa is not susceptible to gonococcal infection.

The natural course of gonorrhea is less well understood in women than in men. Symptoms probably develop in most infected women,[121] but many remain asymptomatic or have only minor symptoms and do not seek medical care.[122] Thus, women with subclinical infection accumulate in the population, and in settings in which sexually active women are routinely screened for subclinical infection up to 80% of women with gonorrhea are likely asymptomatic. Women infected with AHU$^-$ and related auxotypes of *N. gonorrhoeae,* like men, are more likely to have asymptomatic infection and more subtle inflammatory signs.[119]

Most infected women who develop symptoms do so within 10 days. The dominant symptoms are increased vaginal discharge, dysuria (often without urgency or frequency), and intermenstrual bleeding, sometimes triggered by coitus. Abdominal or pelvic pain usually denotes ascending infection, but some women with these symptoms lack evidence of salpingitis at laparoscopy. Physical examination may show purulent or mucopurulent cervical exudate (Fig. 212.13) and other signs of mucopurulent cervicitis, such as edema in a zone of cervical ectopy or endocervical bleeding induced by gentle swabbing[123]; in most infected women, these cervical signs are absent. Purulent discharge can sometimes be expressed from the urethra or the ducts of Bartholin glands.

Rectal Gonococcal Infection

N. gonorrhoeae can be isolated from the rectum in up to 40% of women and a similar proportion of MSM with uncomplicated gonorrhea. The rectum is often the only infected site in MSM. Isolated rectal infection in women is probably more common than traditionally thought; in a large study in Baltimore, 2.75% of 4402 women had gonorrhea detected at the rectum; routine screening of women at this site is, however, not routinely recommended.[124] Rectal infection is acquired both through

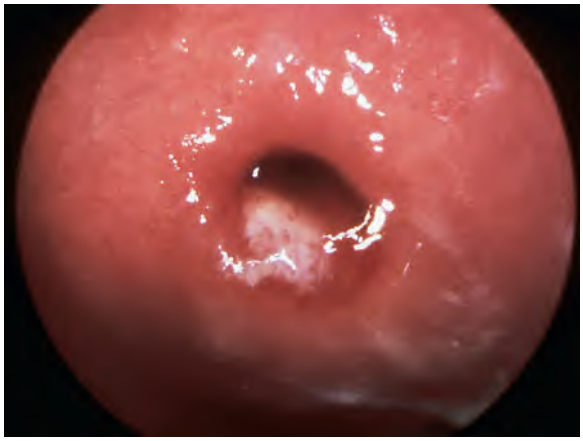


FIG. 212.13 Purulent endocervical exudate in gonococcal cervicitis. *(From Handsfield HH.* Color Atlas and Synopsis of Sexually Transmitted Diseases. *2nd ed. New York; McGraw-Hill; 2001; courtesy King K. Holmes.)*

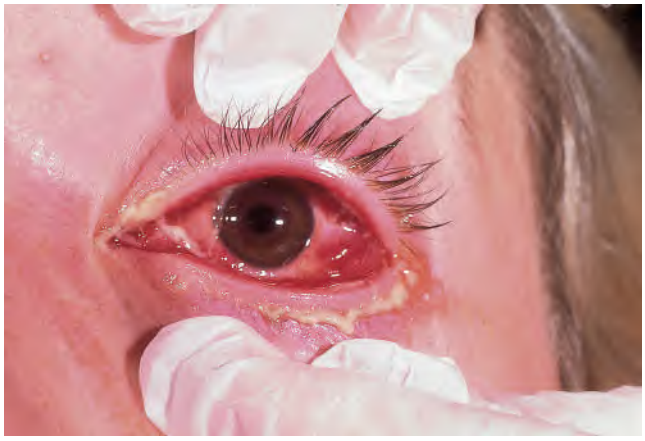


FIG. 212.14 Acute gonococcal conjunctivitis in an adult. *(From Handsfield HH.* Color Atlas and Synopsis of Sexually Transmitted Diseases. *2nd ed. New York; McGraw-Hill; 2001.)*

receptive anal intercourse, which accounts for virtually all cases in men, and perineal contamination with cervicovaginal secretions, which is believed to be responsible for some infections in women. Rectal gonococcal infection is usually asymptomatic, but some patients have acute proctitis manifesting with anal pruritus, tenesmus, purulent discharge, or rectal bleeding. The apparently higher rate of clinically apparent proctitis in men than in women suggests that inoculum size or trauma during anal intercourse may influence the development of symptoms. Anoscopy sometimes reveals mucopurulent exudate and inflammatory changes in the rectal mucosa, but infection with *C. trachomatis,* herpes simplex virus, or other sexually transmitted pathogens can produce the same findings.

Pharyngeal Infection

Pharyngeal gonococcal infection is acquired through receptive oral sex but probably rarely, if ever, by kissing. Acquired more efficiently by fellatio than by cunnilingus, pharyngeal infection can be found in 10% to 20% of heterosexual women with gonorrhea and 10% to 25% of infected MSM, but it is present in only 3% to 7% of heterosexual men with gonorrhea. Almost all pharyngeal infections are asymptomatic, but rare cases may cause overt pharyngitis.

The importance of documenting pharyngeal gonorrhea is debated, and several factors argue against routine screening or diagnostic testing in the general sexually active adult population. Most cases are asymptomatic and resolve spontaneously, and pharyngeal infection is probably less transmissible than rectal or genital gonorrhea. Furthermore, pharyngeal non–culture-based diagnostic assays for identification of *N. gonorrhoeae* have not been cleared by the US Food and Drug Administration (FDA), although they have been validated by numerous health department and several commercial laboratories. On the other hand, pharyngeal infection can be symptomatic and is sometimes the source of transmission to sex partners, especially among MSM,[125] or of systemic dissemination of *N. gonorrhoeae.* Current recommendations are to routinely test for pharyngeal infection in HIV-infected people who report receptive oral sex,[127] and many experts recommend routine pharyngeal screening in MSM given the continued high incidence of disease in this group.[127]

Other Local Manifestations

Gonococcal conjunctivitis in adults is usually seen in people with genital gonorrhea, and most cases probably result from autoinoculation, but some cases may be acquired by other routes, such as orogenital exposure. Gonococcal conjunctivitis is usually painful, with prominent photophobia and copious, purulent exudate (Fig. 212.14), and corneal ulceration can supervene rapidly in the absence of prompt antibiotic therapy. However, some infections are mild, perhaps related to specific gonococcal strains. *N. gonorrhoeae* has been isolated in cases of acute gingivitis, otherwise unexplained oral ulcerations, and intraoral abscesses. Cutaneous abscesses have been described, typically involving the finger or the penile shaft, and probably result from inoculation of preexisting lesions. For example, gonococcal abscess is sometimes the first clinical presentation of a congenitally patent median raphe duct of the penis. Finally, in the absence of neonatal prophylaxis, maternal infection with *N. gonorrhoeae* may result in ophthalmia neonatorum, typically manifesting as purulent, edematous conjunctivitis. In areas without routine neonatal prophylaxis, blindness can result. Infection of other superficial mucosal surfaces can also occur.

Pelvic Inflammatory Disease

PID (see Chapter 111) refers to a spectrum of upper genital tract infections and may occur with or without overt symptoms. When symptomatic, PID manifests with various combinations of endometritis, salpingitis, tubo-ovarian abscess, pelvic peritonitis, and perihepatitis. The immediate and long-term sequelae are a primary impetus for prevention strategies against gonorrhea and chlamydial infection. Infection with *N. gonorrhoeae, C. trachomatis,* perhaps *Mycoplasma genitalium,* and vaginal anaerobes associated with bacterial vaginosis may be involved, often in combination. Adolescent girls are at higher risk than older women, probably because of both innate susceptibility, perhaps due to a high prevalence of cervical ectopy that may facilitate ascending infection, and sexual network factors that confer a higher likelihood of exposure to infected sex partners. Women using anovulatory hormones have been shown to have a lower risk for chlamydial salpingitis but not of gonococcal PID in some studies[128]; however, other analyses have not reported this association.[129]

PID is estimated to occur in 10% to 20% of women with gonorrhea detected at the cervix. In 57 women with gonorrhea but with no clinical signs or symptoms of PID, endometrial biopsies revealed histologic endometritis (termed *subclinical PID* by the authors) in 15 (26%); this was a significantly higher percentage than that seen in women without gonorrhea (11%; $P < .001$).[130]

The most consistent symptom of PID is low abdominal pain, and most women also have symptoms of lower genital tract infection. Gonococcal PID often follows the onset of menses by a few days.[122] Fever, chills, nausea, and vomiting may occur, but most patients lack these manifestations. The primary finding at physical examination is pelvic adnexal tenderness, usually bilateral. Other common findings are uterine fundal tenderness, pain elicited on moving the cervix, and one or more tender adnexal masses. Abdominal examination usually elicits tenderness over the lower quadrants, and signs of peritoneal inflammation are common in severe cases. Many women with PID have bacterial vaginosis, and many have signs of mucopurulent cervicitis. Fever, leukocytosis, and an elevated erythrocyte sedimentation rate or C-reactive protein level are common, but they are absent in approximately one-third of patients with laparoscopically documented PID. In practice, the clinical diagnosis of PID is imprecise.

The proportion of PID cases associated with gonorrhea varies greatly across population groups and with the background rates of gonorrhea and chlamydial infection. In the 1980s, 20% to 40% of cases of PID in most urban areas of the United States were associated with gonorrhea. The presence of cervical gonococcal or chlamydial infection does not exclude fallopian tube infection with other organisms, nor does failure to isolate *N. gonorrhoeae* or *C. trachomatis* definitively exclude their contribution to salpingitis. Along with *M. genitalium,* facultative and anaerobic bacterial microbiota of the vagina may also contribute to acute PID, especially in patients with pelvic abscess or otherwise severe infections.[131]

Infertility resulting from fallopian tube obstruction is the most common serious consequence of PID and occurs in 15% to 20% of women after a single episode and 50% to 80% of those who experience three or more episodes.[132] Infertility may be more common after chlamydial than after gonococcal PID, perhaps because the more acute inflammatory signs associated with gonorrhea bring women to diagnosis and treatment sooner. This pattern was evident in the PID Evaluation and Clinical Health study, which enrolled more than 800 female patients aged 14 to 37 years with symptomatic PID.[133] Despite clinical cure and apparent microbiologic eradication of gonorrhea, as evidenced by lower tract cultures, infertility rates were 13% for participants with *N. gonorrhoeae* identified, 19% for those with *C. trachomatis,* and 22% for those with anaerobic bacteria during a median follow-up period of 35 months.[134] The rate of chronic pelvic pain was 27% among women with gonococcal infection.[135]

Perihepatitis

Acute perihepatitis, or Fitz-Hugh–Curtis syndrome, occurs primarily by direct extension of *N. gonorrhoeae* or *C. trachomatis* from the fallopian tube to the liver capsule and overlying peritoneum. Some cases may result from lymphangitic spread or bacteremic dissemination, which may explain rare cases of apparent perihepatitis in men. Perihepatitis results in abdominal pain, hepatic tenderness, and right upper quadrant peritoneal inflammatory signs. Most cases occur in association with overt PID, but many women lack pelvic symptoms or signs. Perihepatitis should be considered in the differential diagnosis of right upper quadrant pain in young, sexually active women; it is commonly mistaken for acute cholecystitis or viral hepatitis. Laparoscopy may show the characteristic "violin string" adhesions between the liver capsule and the parietal peritoneum.

Gonorrhea in Pregnancy

Gonorrhea during pregnancy is associated with spontaneous abortion, premature labor, early rupture of fetal membranes, and perinatal infant mortality.[136] The clinical manifestations of gonorrhea are unchanged in pregnant women, except that PID and perihepatitis are uncommon after the first trimester, when the products of conception obliterate the uterine cavity. Reports are conflicting as to whether pregnancy is a risk factor for gonococcal bacteremia.

Disseminated Gonococcal Infection

Disseminated gonococcal infection (DGI) results from bacteremic dissemination of *N. gonorrhoeae,* although immune complexes or other indirect immunologic mechanisms may contribute to pathogenesis and symptoms in some cases. Although once estimated to occur in 0.5% to 3% of infected patients, the rate undoubtedly is lower at present because of declining prevalences of gonococcal strains prone to disseminate.[137] However, discrete outbreaks of DGI have recently been documented.[138] Septic arthritis and a characteristic syndrome of polyarthritis and dermatitis are the predominant manifestations, and DGI is a cause of infective arthritis in young adults. Gonococcal endocarditis was common in the preantibiotic era but now is rare. Meningitis, osteomyelitis, septic shock, and acute respiratory distress syndrome are rare manifestations.

Properties of *N. gonorrhoeae* classically associated with dissemination include resistance to the bactericidal action of nonimmune human serum, the AHU$^-$ auxotype, specific Por IA serovars, and marked susceptibility to penicillin—characteristics also associated with asymptomatic genital gonorrhea. However, recent DGI cases have been associated with other auxotype/serovar classes as the prevalence of Por IA/AHU$^-$ gonococci has declined and antibiotic-resistant strains have caused DGI.

Complement deficiency predisposes to gonococcal and meningococcal bacteremia.[139] Up to 13% of patients with DGI have been reported to have complement deficiencies, and patients with repeated episodes of neisserial bacteremia should be tested with an assay for total hemolytic complement activity. Other host factors associated with dissemination include female sex, menstruation, and perhaps pharyngeal gonococcal infection and pregnancy. In approximately half of affected women, symptoms of DGI begin within 7 days of the onset of menses.

The most common presentation of DGI is the arthritis-dermatitis syndrome.[140] During the first few days, most patients experience polyarthralgias that primarily involve the knees, elbows, and more distal joints; the axial skeleton is usually not involved. Physical examination usually shows objective signs of arthritis or tenosynovitis in at least two joints. Asymmetrical involvement of only a few joints helps distinguish DGI from polyarthritis caused by most immune complex–mediated disorders, which typically manifest with symmetrical involvement of many joints. A characteristic dermatitis (Fig. 212.15) is present in approximately 75% of patients and consists of discrete papules and pustules, often with a hemorrhagic component. Hemorrhagic bullae or overtly necrotic lesions that mimic ecthyma gangrenosum are sometimes seen. The lesions usually number 5 to 40 and occur predominantly on the extremities. Fever, systemic toxicity, and polymorphonuclear leukocytosis are common, but they are usually mild and are often absent.

If the infection is not treated, inflammation regresses in most joints and the dermatitis resolves, but overt septic arthritis supervenes in one or two joints, most commonly the knee, ankle, elbow, or wrist, although any joint may be involved. Septic gonococcal arthritis develops in some patients without prior polyarthritis or dermatitis; in the absence of the characteristic dermatitis or overt genital infection, it is clinically indistinguishable from septic arthritis of any etiology.

During the arthritis-dermatitis stage, gonococci often can be recovered through blood culture, but synovial fluid, if it can be obtained, usually contains fewer than 20,000 leukocytes/mm^2 and is sterile. Gonococci can often be seen by means of immunochemical methods in biopsy specimens of skin lesions, but cultures are generally sterile. In septic gonococcal arthritis, synovial fluid usually contains more than 50,000 leukocytes/mm^2 and culture is often positive, but at this stage blood cultures are usually negative.

Gonococcal bacteremia is often intermittent, so a minimum of three blood cultures should be obtained when DGI is suspected. Synovial fluid should be cultured if a specimen can be obtained. However, only approximately half of patients with DGI have positive cultures of blood or synovial fluid. Although uncommonly positive, Gram-stained smears and cultures of pustular skin lesions are simple to perform and should be obtained. *N. gonorrhoeae* can be recovered from a mucosal site in at least 80% of patients, so the urethra or endocervix, the rectum, and the pharynx should be tested regardless of symptoms or exposure history, and patients' sex partners should be examined and tested. Nucleic acid amplification tests (NAATs), including polymerase chain reaction assay, are more sensitive than culture in detecting gonococci in synovial fluid and have been applied to skin lesions of DGI with success.[141] The diagnosis of DGI is confirmed if *N. gonorrhoeae* is identified in a nonmucosal specimen such as blood, synovial fluid, or a skin lesion, and the diagnosis is probable if infection is documented at a mucosal site or in a sex partner of a patient with a typical clinical syndrome.

The differential diagnosis of the gonococcal arthritis-dermatitis syndrome includes meningococcemia, septic arthritis due to other pyogenic bacteria, and the entire range of inflammatory arthritis. Reactive arthritis is easily confused with DGI because it is common in sexually active young adults and is associated with urethritis, cervicitis, and skin lesions that sometimes have a pustular component. Usually, careful clinical and microbiologic assessment readily differentiates these disorders, but a trial of antibiotic therapy may be required.

Gonococcal endocarditis, which usually involves the aortic valve, is a rare but serious manifestation that occurs in an estimated 1% or 2% of patients with DGI.[142] Although often associated with the arthritis-dermatitis syndrome, endocarditis may be the sole manifestation of DGI. In the preantibiotic era, median survival was 6 to 8 weeks, reflecting

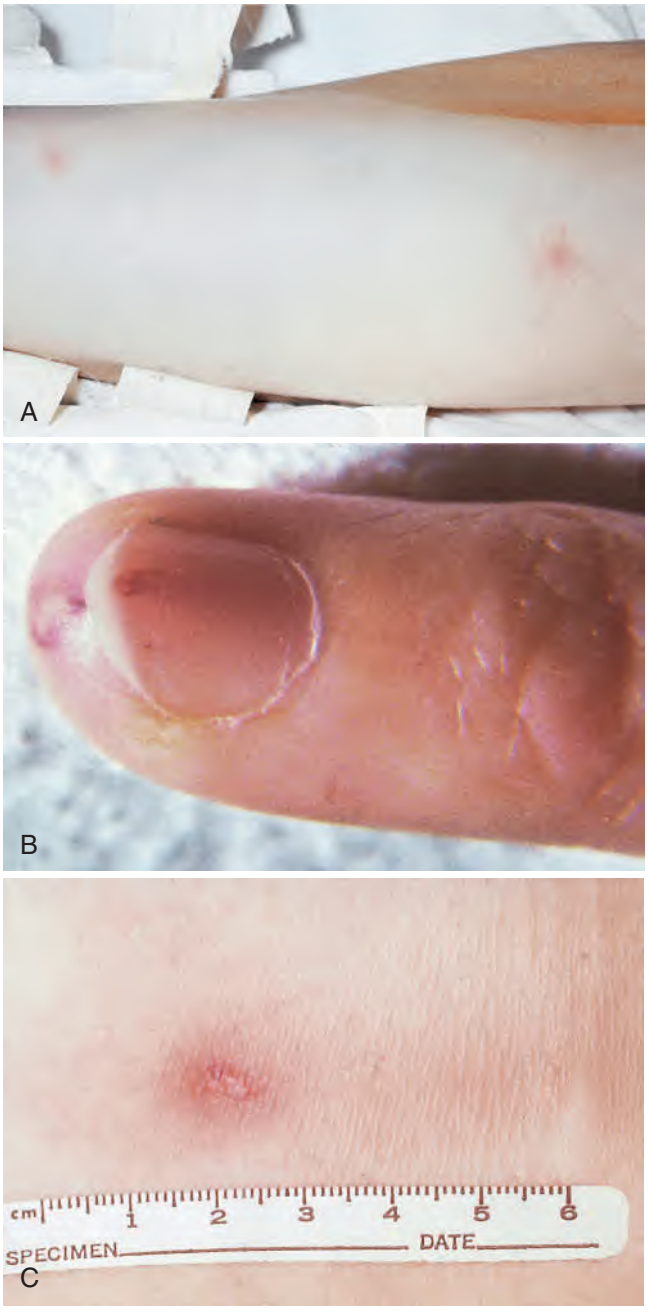


FIG. 212.15 Cutaneous lesions in disseminated gonococcal infection. (A) Early papular lesions. (B) Pustular lesion associated with subungual hemorrhage. (C) Ulcerated pustular lesion.

a typical rate of valve destruction between that of acute staphylococcal or pneumococcal endocarditis and subacute endocarditis caused by viridans streptococci.

Neonatal and Pediatric Infections

Gonococcal conjunctivitis of the newborn (ophthalmia neonatorum) is the most common clinically recognized manifestation of neonatal infection[143]; it was once a common cause of blindness in the United States and may remain so in some developing countries. Prophylaxis by means of instillation of antibiotics into the conjunctivae soon after delivery is highly effective, although occasional failures occur. The most important preventive measure is routine screening and treatment of pregnant women for gonorrhea before term. The diagnosis of gonococcal ophthalmia should be suspected clinically when acute conjunctivitis develops within a few days of delivery and is confirmed by identification of gonococci in conjunctival secretions. Systemic illness with septicemia and arthritis can also develop in neonates exposed to gonorrhea, but these are rare. *N. gonorrhoeae* can often be recovered from the gastric aspirates of infants born to infected mothers, but many cases probably reflect transient colonization rather than clinically important infection.

Rectal gonococcal infection is sometimes seen in neonates, but vaginal infection is uncommon because the neonatal vaginal mucosa is well estrogenized by circulating maternal hormone. Purulent vaginitis is the primary manifestation of gonorrhea in prepubertal girls after the neonatal period, but otherwise the clinical manifestations of gonorrhea in children are not materially different from those in adults. After the neonatal period until 1 year of age, most cases in children appear to be acquired nonsexually from an infected parent, usually in a setting of poor hygiene. After 1 year of age, most cases are acquired by sexual abuse. Nonsexual transmission of ocular infection occasionally occurs in young children in tropical settings.

DIAGNOSIS

Isolation of *N. gonorrhoeae* is the historical standard for diagnosis. Culture is inexpensive and reasonably sensitive and preserves an isolate for antimicrobial susceptibility testing when clinically indicated or for surveillance. Culture is widely considered the only appropriate test in forensic settings—for example, in testing people who have been sexually assaulted or children with suspected gonorrhea. Nevertheless, NAATs have supplanted culture in most settings, owing to convenient specimen management and their usefulness in testing urine and self-obtained vaginal swabs. Nonamplified DNA probe tests remain in use despite their lower sensitivity compared with culture or NAATs. Microscopy of gram-stained smears is effective in the diagnosis of symptomatic urethritis in men but has marginal usefulness in other settings. There is no clinically useful serologic test.

Culture

A single culture on antibiotic-containing selective medium, such as modified Thayer-Martin agar, has a sensitivity of 95% or more for urethral specimens from men with symptomatic urethritis and 80% to 90% for endocervical infection in women. Results may vary depending on the quality of the medium and the adequacy of the clinical specimen. Simultaneous inoculation of both selective and nonselective media maximizes sensitivity for cervical infection but is impractical in most settings. Normally, sterile clinical specimens, such as blood, synovial fluid, and cerebrospinal fluid, should be inoculated onto enriched chocolate agar or another nonselective medium in addition to broth medium and should also be tested with NAATs, but no published studies have systematically analyzed the detection of *N. gonorrhoeae* in broth cultures with the automated techniques now used by most clinical laboratories.

Up to 5% of gram-negative, oxidase-positive diplococci isolated from the genital tract and substantially greater proportions of those from the rectum and pharynx are in fact *N. meningitidis*.[144] Some of these cases are associated with urethritis, cervicitis, or proctitis that is clinically indistinguishable from gonorrhea.[145,146] Therefore, speciation of *Neisseria* isolates is desirable for positive genital cultures and should be routine for pharyngeal and rectal specimens. Available NAATs for *N. gonorrhoeae* reliably exclude meningococcal infection. Antimicrobial susceptibility testing of *N. gonorrhoeae* is not generally recommended except when one is conducting surveillance for antimicrobial resistance, but it should be routine after treatment failure and in cases of disseminated infection.

Nucleic Acid Amplification Tests

Currently available NAATs for *N. gonorrhoeae* use several related technologies, including transcription-mediated amplification, polymerase chain reaction assay, and the DNA strand displacement assay. The NAATs have largely replaced culture in most settings in which people are screened for asymptomatic genital infection and, in particular, in which assays with combined targets are used for gonorrhea and chlamydial infection (e.g., the APTIMA Combo 2 assay [Hologic]). These tests are not significantly more sensitive than culture for detecting *N. gonorrhoeae* in cervical or urethral specimens (unlike the diagnosis of *C. trachomatis,* for which the NAATs are more sensitive than all other tests). However,

the NAATs for *N. gonorrhoeae* have specificities greater than 99%, offer important advantages in specimen management, and retain sensitivity when used to test voided urine or self-collected vaginal swabs, an important advantage for laboratory screening in many settings. The transcription-mediated amplification test has a positive predictive value of at least 95% in women in settings in which gonorrhea prevalence is as low as 0.5%, indicating that false-positive results are rare even when low-prevalence populations are screened.[147] Concern has been expressed that persisting DNA may cause false-positive results when NAATs are used to test patients soon after treatment, but NAAT results for *N. gonorrhoeae* become negative within 1 to 2 weeks of successful treatment.[148] In fact, the recommendation to obtain a test of cure at 1 week if indicated includes NAAT testing and assumes that the results of these assays will rapidly become negative for *N. gonorrhoeae.*[107] The APTIMA test (and possibly others, although fewer data exist at this point) for *N. gonorrhoeae* is also reliable for diagnosis of rectal and pharyngeal infection.[149,150] Although these assays have not been cleared by the FDA for routine use, the CDC recommends their use as long as laboratories are Clinical Laboratory Improvement Amendments (CLIA) compliant with regard to test modifications, before reporting the results for patient management.[151] These observations may not extend to other NAATs; the Probe-Tec assay demonstrated low positive predictive value when applied to pharyngeal and rectal specimens in a community-based sample of MSM.[152]

Gram-Stained Smears

The Gram stain has been used for more than a century to identify gonococci in clinical specimens; methylene blue and other dyes have also been used. Microscopy findings of Gram-stained smears are positive if gram-negative diplococci of typical morphology are observed in association with neutrophils (Fig. 212.16), negative if no such organisms are seen, and equivocal if typical morphotypes are not associated with neutrophils or if morphologically atypical organisms are seen. Nonpathogenic members of the Neisseriaceae are not usually associated with leukocytes. In men with symptomatic urethritis, microscopy is at least 95% sensitive and is highly specific for diagnosis of gonorrhea, but microscopy is less useful for other anatomic sites. The sensitivity approximates 50% for asymptomatic urethral infection in men and for cervical or rectal infection; the specificity is said to be high, but this likely is true only for highly experienced observers. Microscopy is both insensitive and nonspecific for detection of pharyngeal gonococcal infection and is not recommended.

Other Diagnostic Methods

Nonamplified DNA probe tests are less sensitive than culture or NAATs, and they are not useful in the diagnosis of rectal or pharyngeal infection or for testing urine. However, these assays are inexpensive and are offered by many laboratories in combination with assays for *C. trachomatis.* Immunochemical detection of gonococcal antigens, such as with enzyme immunoassays with polyclonal antigonococcal antibodies and fluorescein-conjugated monoclonal antibodies for direct fluorescence microscopy, now is little used.

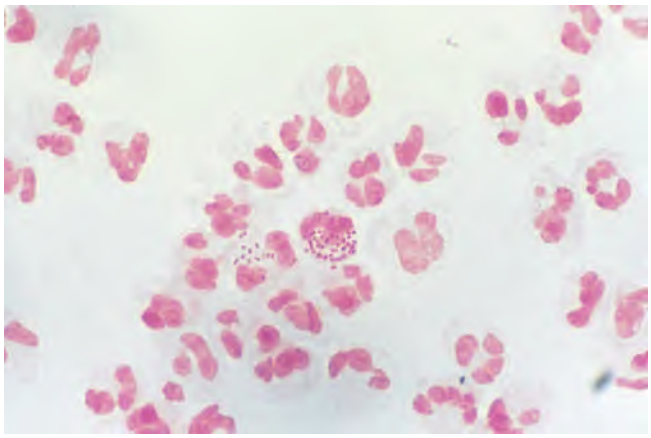


FIG. 212.16 Gram-stained smear of urethral exudates showing intracellular gram-negative diplococci that are characteristic of gonorrhea.

THERAPY

The antimicrobial susceptibility of *N. gonorrhoeae* is labile, varies greatly across geographic areas and populations, and fluctuates over time. Clinical treatment decisions almost invariably are made without knowledge of the antimicrobial susceptibility of the infecting strain. Therefore, when possible, the regimens for routine treatment of gonorrhea should have efficacies that approach 100%, regardless of the distribution of sensitive and resistant strains of *N. gonorrhoeae* in the community, and treatments with efficacies less than 95% should be avoided.[153] Other factors that influence therapeutic decisions for gonorrhea are the pharmacokinetic characteristics of the agent, its efficacy in complicated versus uncomplicated infection, differential efficacy at various anatomic sites of infection, toxicity, availability, convenience of administration, and cost.[154] An additional consideration is the potential efficacy of an agent for concurrent infection. The most common coexisting pathogen in people with gonorrhea is *C. trachomatis,* and many studies during 3 decades have demonstrated consistent results: 15% to 25% of heterosexual men, 10% to 15% of MSM, and 35% to 50% of women with gonorrhea have concurrent chlamydial infections.[155] The treatment of gonorrhea has been greatly facilitated by the excellent efficacy of single-dose regimens; however, with increasing resistance to several classes of antibiotics (detailed later), this approach has been reevaluated. Gonorrhea in pregnant women responds to the recommended regimens.

Uncomplicated Gonorrhea in Adults
Initial Single-Dose Treatment

Recommendations for the treatment of uncomplicated gonorrhea, modified from those published in the 2015 Sexually Transmitted Diseases Treatment Guidelines from the CDC, are summarized in Table 212.2.[153,154]

TABLE 212.2 Options for the Treatment of Gonorrhea

Uncomplicated Infection of the Cervix, Urethra, and Rectum
Ceftriaxone, 250 mg IM single dose *and* Azithromycin, 1 g PO single dose
Infection of the Pharynx
Ceftriaxone, 250 mg IM single dose *and* Azithromycin, 1 g PO single dose
Conjunctivitis (Not Ophthalmia Neonatorum)
Ceftriaxone, 1 g IM single dose
Disseminated Gonococcal Infection
Ceftriaxone, 1 g IM or IV every 24 hours for 24–48 hours[b] after improvement, with switch to oral therapy for completion of 1 week total antibiotic therapy, including cefixime, 400 mg PO twice daily
Meningitis and Endocarditis
Ceftriaxone, 1–2 g IV every 12 hours for 10–14 days (meningitis) or ≥4 weeks (endocarditis)
Ophthalmia Neonatorum
Ceftriaxone, 25–50 mg/kg IV or IM in a single dose, not to exceed 125 mg[c]

[a]Patients should abstain from sex for 1 week after single-dose treatment. Test of cure for pharyngeal infection is recommended at 14 days if the ceftriaxone regimen is not used.
[b]Ceftriaxone administered IM may be reconstituted in 1% lidocaine solution to minimize injection pain. Alternative parenteral regimens include cefotaxime, ceftizoxime, and spectinomycin. See www.cdc.gov/std/treatment for specific regimens.
[c]Topical antibiotic therapy alone is inadequate for treatment of ophthalmia neonatorum.
Modified from Centers for Disease Control and Prevention. Sexually transmitted diseases treatment guidelines, 2015. MMWR Morb Mortal Wkly Rep. *2015;64:60–68; and www.cdc.gov/std/treatment.*

A single 250-mg intramuscular dose of ceftriaxone is recommended for infection of all mucosal sites, with cure rates that exceed 98% for urethral, cervical, and rectal gonorrhea and 90% for pharyngeal infection; this should be combined with azithromycin given as a single dose of 1 g orally. The rationale for giving a second drug with a different mechanism of action rather than the primary treatment is that it may reduce selection pressure for antimicrobial resistance in *N. gonorrhoeae*. However, as noted earlier, increasing rates of resistance to azithromycin may soon compromise this approach.[116,117] Hypothetically, the worldwide spread of fluoroquinolone-resistant gonococci may have been slower than the dissemination of penicillinase-producing strains 25 years earlier in part because penicillin monotherapy was the rule in the 1970s, whereas antichlamydial cotherapy was the norm in the 1990s. Because of increasing resistance to the cephalosporins, neither of the widely available oral third-generation cephalosporins—cefixime and cefpodoxime—is recommended for primary therapy. Of note, allergic cross-reactions between penicillin and third-generation cephalosporins appear to be uncommon, and penicillin-allergic people can often be treated for gonorrhea with a cephalosporin.[156] Both ceftriaxone and azithromycin are safe and effective in pregnant women.

Unfortunately, owing to the relentless worldwide increase in the resistance of *N. gonorrhoeae* strains to fluoroquinolones, these agents now have limited clinical usefulness. Increasingly widespread fluoroquinolone resistance in the United States led the CDC to recommend against the use of these agents to treat gonorrhea in MSM in 2004 and, in August 2007, against routine use for all cases of gonorrhea. Nonetheless, fluoroquinolones are highly effective against infection due to susceptible gonococci, with cure rates of at least 98% for all anatomic sites. Although their general use should be discouraged, these drugs retain a useful role in patients in whom no other option is feasible, such as those with absolute contraindications to cephalosporin; however, as noted previously, allergic cross-reactions between penicillin and ceftriaxone appear to be uncommon, and penicillin-allergic people often can be treated for gonorrhea with ceftriaxone. When a fluoroquinolone is the only feasible option for gonorrhea treatment, a culture isolate should be obtained so that susceptibility testing can be performed and for reference in the event of treatment failure.

In one clinical trial, treatment of uncomplicated urogenital gonorrhea with dual therapy consisting of oral azithromycin (2 g) and either a single oral dose of gemifloxacin (320 mg) or a single intramuscular dose of gentamicin (240 mg) was associated with cure rates of 99.5%, and 100%, respectively.[157] Few infections in this trial occurred at rectal and pharyngeal sites, limiting the reliability of cure estimates at these sites; regardless, if available, either of these regimens may be considered alternative therapies, although gastrointestinal side effects limited tolerability.

Spectinomycin, 2 g intramuscularly, is effective for genital and rectal gonorrhea, but it is ineffective for pharyngeal infection and is currently not available in the United States; its eventual availability is uncertain. Its sole indication is the treatment of pregnant women with a history of rapid-onset allergic reactions to penicillin or documented cephalosporin allergy—a rare confluence of events. Other options to treat such women are limited and have not been studied; these include desensitization to recommended agents or use of alternative drugs with test of cure.

Follow-Up of Patients Treated for Uncomplicated Gonorrhea

Failure of treatment of infections with organisms that are susceptible to recommended regimens is uncommon. Retesting should be performed to evaluate persistence of symptoms at any infected site; ideally, this should be performed with culture to facilitate assessment for antimicrobial resistance. Documentation of cure should be performed with retesting for pharyngeal infections treated with a regimen other than ceftriaxone. In this case, test of cure using NAATs or culture should be obtained 14 days after treatment was provided. It is important to realize, however, that most patients with gonorrhea remain at risk for repeat infection even if initial treatment was successful; studies have found recurrent gonorrhea in 10% to 15% of people retested 1 to 4 months after treatment.[158–160] Accordingly, retesting for recurrent or persistent infection, or rescreening, should be done routinely 3 or 4 months after treatment.

TABLE 212.3 Recommended Treatment of Acute Pelvic Inflammatory Disease

Hospitalized Patients

Regimen A

Cefotetan, 2 g IV q12h, or cefoxitin, 2 g IV q6h
plus
Doxycycline, 100 mg IV or PO q12h
Continue both drugs IV for 24 hours after the patient substantially improves, then continue doxycycline, 100 mg PO bid, to complete 14 days total therapy. Either clindamycin or metronidazole may be added to the oral regimen if tubo-ovarian abscess is suspected.
or

Regimen B

Clindamycin, 900 mg IV q8h
plus
Gentamicin, 2 mg/kg IV once, followed by 1.5 mg/kg q8h[a]
Continue both drugs IV for 24 hours after the patient substantially improves, then continue doxycycline, 100 mg PO twice daily, or clindamycin, 450 mg PO four times daily, to complete 14 days total therapy. Clindamycin may be preferable when tubo-ovarian abscess is suspected.

Outpatients

Single-dose cefoxitin, 2 g IM, plus probenecid, 1 g PO; or ceftriaxone, 250 mg IM; or other parenteral third-generation cephalosporin (e.g., ceftizoxime or cefotaxime)
plus
Doxycycline, 100 mg PO bid for 14 days
with or without
Metronidazole, 500 mg PO bid for 14 days

[a]Single daily dosing may be substituted.
Modified from Centers for Disease Control and Prevention. Sexually transmitted diseases treatment guidelines, 2015. MMWR Morb Mortal Wkly Rep. *2015;64:60–68; and www.cdc.gov/std/treatment.*

Rescreening is also indicated for men and women with chlamydial infection. Testing urine or self-collected vaginal specimens with NAATs can be accomplished in asymptomatic patients without a clinical examination or, if using mailed specimens, without a clinic visit.

Pelvic Inflammatory Disease

The treatment of PID is addressed in detail in Chapter 111. In individual patients, the specific pathogens responsible for ascending genital infection usually are not known and the recommendations for initial treatment of acute PID are similar regardless of whether the initial infection is due to *N. gonorrhoeae, C. trachomatis,* or other pathogens. Guidelines for the treatment of PID are summarized in Table 212.3. The main regimen for outpatient therapy is single-dose treatment with ceftriaxone or with cefoxitin plus probenecid, followed by oral therapy with doxycycline, 100 mg twice a day orally, and metronidazole, 500 mg twice a day orally for 14 days. In a well-designed study, this regimen (without metronidazole) was equally as effective as the equivalent inpatient parenteral regimen both in short-term resolution and in preservation of fertility after a 3-year follow-up period.[134] The use of azithromycin for treatment of PID has been controversial and is not currently recommended. The CDC guidelines stipulate that fluoroquinolone may be considered as the antibiotic for gonorrhea coverage in PID if the community prevalence and individual risk for fluoroquinolone-resistant gonorrhea are low, and the guidelines recommend performing diagnostic testing with appropriate management according to the results. In addition, in many communities gonorrhea is now a relatively uncommon cause of PID, reducing concern about antibiotic resistance.

Regardless of the initial treatment, close follow-up is indicated. Clinical progression or absence of improvement within 3 days is an indication for diagnostic imaging and hospitalization of the patient for parenteral therapy and possible laparoscopy to confirm the diagnosis and obtain intraabdominal culture specimens to facilitate selection of improved, parenteral antimicrobial therapy. The patient's sex partner(s) should be tested and treated for chlamydial infection and gonorrhea, unless both infections can be reliably excluded in the index patient.

Acute Epididymitis

The treatment of acute epididymitis is addressed in Chapter 112. Most cases in young adults are due to *C. trachomatis* or *N. gonorrhoeae,* but coliforms and other urinary tract pathogens are common causes in older men or after urinary tract instrumentation and perhaps in men who participate in unprotected insertive anal intercourse. Most cases can be managed on an outpatient basis with ceftriaxone, 250 mg intramuscularly, plus 10 days of treatment with doxycycline, 100 mg twice daily. Ofloxacin, 400 mg orally twice daily, or levofloxacin, 500 mg daily, for 10 days, provides better coverage for coliforms and nonsexually transmitted pathogens and is effective against chlamydial infection but should be avoided if gonorrhea is suspected.

Disseminated Gonococcal Infection

Few data are available on the treatment of patients with DGI since the evolution and spread of gonococci with high levels of antibiotic resistance, and all recommendations are empirical. Antimicrobial susceptibility testing should be routinely used to guide therapy in case the initial response to empirical treatment is suboptimal. Patients with the gonococcal arthritis-dermatitis syndrome should be treated initially with ceftriaxone, 1 g once daily, given either intramuscularly or intravenously.[153] Most patients without complications can be treated as outpatients. Equivalent doses of other third-generation cephalosporins undoubtedly would be effective and are recommended as options. No reported studies have systematically evaluated the fluoroquinolones, but these drugs undoubtedly are effective if the organism is susceptible. After clinical improvement begins, treatment of patients without septic arthritis or other complications may be switched to an oral cephalosporin (e.g., cefixime 400 mg twice daily) or a fluoroquinolone (e.g., levofloxacin 500 mg daily) to complete 7 to 10 days of total therapy. Patients with gonococcal endocarditis should receive 4 weeks of parenteral therapy, usually starting with ceftriaxone or an equivalent cephalosporin and later modifying the regimen if dictated by the results of antimicrobial susceptibility testing. Meningitis should be treated with a 10- to 14-day course of ceftriaxone.

Gonorrhea in Children

Relatively few cases of gonorrhea are seen in children, and treatment has not been well studied; most treatment recommendations are extrapolated from those for adults. Uncomplicated infections in neonates and older children should normally be treated with ceftriaxone in a single intramuscular dose of 25 to 50 mg/kg, up to 125 mg.[153] Little is known about the prevalence of chlamydial infection in pediatric patients with gonorrhea, and the usual practice is to perform a test for *C. trachomatis* and withhold specific treatment unless infection is diagnosed. DGI and gonococcal conjunctivitis in children are treated with 7 to 10 days of ceftriaxone, 25 to 50 mg/kg/day intramuscularly or intravenously, or with an equivalent regimen of another third-generation cephalosporin. Continuous irrigation of the conjunctivae with physiologic saline solution is often used in gonococcal conjunctivitis, but topical antibiotics probably offer no additional benefit.

Management of Sex Partners

Management of sex partners is an integral part of treating patients with gonorrhea and other STDs because failure to ensure that the partner is treated risks reinfection of the patient and fosters continued transmission. Few state or local health departments in the United States provide direct assistance in contacting or managing the partners of people with gonorrhea or chlamydial infection diagnosed outside public clinics (and often not there); the physician and patient should work together to this end.

Ideally, the sex partners of people with gonorrhea or chlamydial infection should be examined, tested for both infections and other common STDs, and counseled on prevention. Unfortunately, success in bringing partners to clinical care is low, and in most settings treatment can be documented for less than half the partners of infected people.[161] Studies suggest that this proportion can be increased and the rate of reinfection in index patients reduced by arranging for treatment without examination—that is, by giving the index patient an antibiotic for the partner or a prescription in the partner's name or by calling in a prescription directly for the partner. Collectively, these strategies have been termed *expedited partner therapy* (EPT).[162] Several states with formerly restrictive laws or regulations have modified them to permit EPT, and others are considering such steps. The CDC provides up-to-date reports of the legal status of EPT throughout the United States at www.cdc.gov/std/ept. Clinicians should routinely use EPT for the partners of people with gonorrhea or chlamydial infection, to the extent permitted by regional laws and regulations, whenever success is not ensured in personal evaluation of the partners. EPT for gonorrhea can be accomplished with single-dose oral therapy active against both *N. gonorrhoeae* (e.g., cefixime) and *C. trachomatis* (azithromycin); for the partners of patients with chlamydial infection alone, azithromycin is appropriate.

PREVENTION AND CONTROL

Public Health Strategies

Screening of sexually active people is a mainstay of public health strategies to prevent gonorrhea and the other treatable bacterial STDs, and it is largely responsible for the dramatic declines observed in the incidence of gonorrhea nationwide after the mid-1970s. Women at risk who undergo routine pelvic examinations should be tested for *N. gonorrhoeae* and *C. trachomatis* according to published guidelines where available. The US Preventive Services Task Force recommends routine annual testing for gonorrhea in sexually active women younger than 25 years and for older women with specific risks.[163] Some authors have used more refined epidemiologic analyses to help narrow these relatively broad recommendations for screening and have suggested that local gonorrhea prevalence rates may be especially useful.[164] When pelvic examination is otherwise not indicated, urine or a self-collected vaginal swab can be tested with NAATs. Universal screening of all heterosexual men and women usually is not cost-effective except in certain settings, such as public STD clinics. In screening women or heterosexual men, a pharyngeal culture should be obtained if symptoms of pharyngitis are present, and it may be warranted if the patient has performed oral sex on a person known to have genital gonorrhea.

Although asymptomatic cervical gonorrhea has been recognized for some time, of increasing concern is the underappreciated prevalence of asymptomatic gonorrhea in some populations, particularly rectal and pharyngeal infection in MSM. Sexually active MSM should be tested routinely for both gonococcal and chlamydial infection, depending on the anatomic sites exposed. Furthermore, it was shown more than a decade ago that urethritis increases HIV viral load in semen, and treatment concomitantly lowers levels of HIV RNA.[165] Thus, prompt diagnosis and treatment of urethritis along with routine periodic screening at all exposed anatomic sites is recommended for all HIV-infected people at initial evaluation and thereafter at least every 12 months and more frequently for those at higher risk.[127] Among asymptomatic MSM, the highest yield will be achieved by testing the rectum if receptive anal intercourse is reported, urethra if insertive anal sex is reported, and pharynx if receptive oral sex is reported.

Public health control measures for gonorrhea and chlamydial infection also include routine rescreening and treatment of patients' sex partners (see previous discussion). Other elements include appropriate diagnostic testing in people with compatible clinical syndromes, adherence to recommended treatment regimens, and periodic testing of gonococcal isolates to monitor trends in antimicrobial resistance. Reporting of cases by health care providers, preferably supplemented by direct reporting by laboratories of people with positive test results, is important for epidemiologic monitoring and to facilitate targeted control efforts. Public education and personal counseling, in an effort to encourage healthy sexual behavior and the use of barrier contraceptives, is central to the control of gonorrhea and all STDs. Finally, all people with a newly acquired STD should routinely be tested for other common infections. For example, screening tests for gonorrhea, chlamydial infection, syphilis, and HIV should be undertaken in people diagnosed with any STD; women with STDs should undergo cervical cytology and diagnostic testing for vaginal infections.

Condoms and Microbicides

Properly used male condoms provide a high degree of protection against transmission or acquisition of gonorrhea, chlamydial infection, HIV, and other STDs transmitted by infected secretions.[153] Methodologic

limitations of most studies that have attempted to estimate the magnitude of condom effectiveness tend to result in an underestimate of protection. Nevertheless, a well-designed systematic review reported that most studies found condom use to be associated with reduced risk for both gonorrhea and chlamydia.[166] The female condom likely provides comparable protection against gonococcal infection.[167] Logically, diaphragms and cervical caps might be expected to offer some protection against gonorrhea. Although a randomized controlled cohort study in South Africa failed to show a protective effect against HIV, a subset analysis revealed that women who reported consistent diaphragm use experienced a significant reduction in incident gonococcal infection (relative hazard, 0.61; 95% CI, 0.41–0.91).[168] The spermicide nonoxynol-9 provides no significant protection against gonorrhea or chlamydial infection and is associated with increased risk for vulvovaginal candidiasis and bacterial urinary tract infection and increased risk for HIV acquisition in a large trial.[169] Although spermicidal preparations with nonoxynol-9 enhance the contraceptive efficacy of barrier methods, they should be avoided by people at high risk for STDs, as should condoms packaged with lubricant containing nonoxynol-9. Research is underway to identify alternative microbicides for vaginal or rectal use. No evidence has been presented that such time-honored measures as washing, urinating, or douching after exposure materially reduce the risk for gonorrhea or any other STD.

Other Prevention Strategies

The development of a vaccine to prevent gonorrhea is a high research priority, but an experimental vaccine containing purified gonococcal pili conferred only partial protection against experimental infection with the homologous strain of *N. gonorrhoeae* and no protection from heterologous challenge.[170] The extraordinary degree of antigenic variability in pili, Opa proteins, and LOS, both at the community level and during the course of each infection, presents formidable barriers to developing a gonorrhea vaccine based on these antigens. More antigenically stable proteins, including porin, are under investigation as possible vaccine candidates, but success probably lies in the distant future. Most intriguing is a recent study that used a retrospective case-control approach of patients at sexual health clinics in New Zealand who were eligible to have received an outer membrane vesicle meningococcal B vaccine (MeNZB) in the 1980s. Vaccinated individuals were 31% significantly less likely to have gonorrhea diagnosed subsequently ($P < .001$), suggesting a protective effect and providing a potential pathway for effective vaccine development.[171]

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The complete reference list is available online at Expert Consult.

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213 *Moraxella catarrhalis, Kingella*, and Other Gram-Negative Cocci

Timothy F. Murphy

SHORT VIEW SUMMARY

Definition
- *Moraxella catarrhalis* is a gram-negative diplococcus that colonizes the upper respiratory tract of children.
- It is a common cause of otitis media in children and exacerbations of chronic obstructive pulmonary disease (COPD) in adults.
- *Kingella kingae* is an increasingly recognized pathogen that causes osteoarticular infections and bacteremia in children 6 months to 4 years of age and endocarditis in all ages.

Epidemiology
- *M. catarrhalis* is recovered exclusively from humans.
- Nasopharyngeal colonization is common in infancy and childhood and decreases with age.
- *K. kingae* colonizes the oropharynx of children and is more often isolated from those who attend daycare centers. Infections occur sporadically, but occasional clusters are seen among children in close contact.

Microbiology
- *M. catarrhalis* is easily overlooked in culture because it is phenotypically identical to *Neisseria* in Gram stain and on culture plates.
- *M. catarrhalis* produces oxidase, catalase, and DNase, which are used for speciation.
- *K. kingae* is difficult to isolate and to identify, growing more readily in liquid culture than standard solid agar media.

Diagnosis
- An etiologic diagnosis of the most common clinical manifestations of *M. catarrhalis* (otitis media in children and exacerbations of COPD in adults) is not made routinely in clinical practice. Rather, these infections are usually treated empirically, based on a presumptive diagnosis using clinical manifestations.
- A diagnosis of *K. kingae* infections is made by recovering the organism in culture from normally sterile body fluid, including blood and joint fluid aspirate. Difficulties in isolation can lead to an incorrect diagnosis of culture-negative bone or joint infection. Polymerase chain reaction–based assays are more sensitive than culture in detecting the organism in joint fluid.

Therapy
- Most isolates of *M. catarrhalis* produce β-lactamase and are thus resistant to ampicillin. Isolates are susceptible to amoxicillin/clavulanic acid, macrolides, fluoroquinolones, and extended-spectrum cephalosporins.
- Otitis media and exacerbations of COPD are treated with oral antimicrobial agents, with the choice generally guided by expert guidelines.
- *K. kingae* isolates are usually susceptible to penicillins, cephalosporins, fluoroquinolones, aminoglycosides, trimethoprim-sulfamethoxazole, and macrolides, although a plasmid-encoded β-lactamase has been found in a few strains.

Prevention
- No vaccines are available currently, but research is in progress to develop vaccines to prevent *M. catarrhalis* infections, including otitis media and exacerbations of COPD.
- Interrupting outbreaks of *K. kingae* osteoarticular infections in daycare centers by prophylactic antibiotics has met with mixed success.

Over the past 3 decades *Moraxella (Branhamella) catarrhalis* has emerged as an important and common human respiratory tract pathogen. In this chapter *M. catarrhalis* is discussed. In addition, *Kingella* and other gram-negative cocci, including *Neisseria* other than *N. meningitidis* and *N. gonorrhoeae*, and other *Moraxella*, which are less common causes of human infection, are considered. *Acinetobacter* is discussed in Chapter 222, and *Oligella* is discussed in Chapter 236. *N. meningitidis* and *N. gonorrhoeae* are discussed in Chapters 211 and 212, respectively.

MORAXELLA CATARRHALIS

History

M. catarrhalis has an interesting and checkered taxonomic history. The bacterium was first described a century ago and was suspected by Sir William Osler to be the cause of his own terminal pneumonia.[1] After having been initially named *Micrococcus catarrhalis*, the organism's name was subsequently changed to *Neisseria catarrhalis* because of its similarities in phenotype and ecologic niche to *Neisseria* spp. In 1970 it was transferred to the new genus *Branhamella* on the basis of differences in fatty-acid content and DNA hybridization studies, compared with other Neisseriaceae.[2] The name *Moraxella catarrhalis* was subsequently proposed, and this is the most widely accepted name at this time.

For most of the last century *M. catarrhalis* was regarded as an upper respiratory tract commensal. However, since the late 1970s, investigators from many centers have accumulated compelling evidence that *M. catarrhalis* is an important and common respiratory tract pathogen in humans.[3–9]

Microbiology

Current taxonomic classification schemes include three genera in the family Moraxellaceae—*Moraxella, Acinetobacter*, and *Psychrobacter. M. catarrhalis* is a gram-negative diplococcus that is indistinguishable from *Neisseria* by Gram stain. The organism grows well on blood agar, chocolate agar, and a variety of media. *M. catarrhalis* are difficult to distinguish from *Neisseria* by colony morphology, particularly after overnight growth on agar plates. After 48 hours of growth, *M. catarrhalis* colonies tend to be larger than *Neisseria* and take on a pink color. In addition, colonies display the hockey puck sign by sliding along the surface of the agar when pushed. Because samples from the respiratory tract frequently contain *Neisseria*, suspicious colonies should be tested for the possibility that they are *M. catarrhalis*. The similarity in colony morphology between commensal *Neisseria* and *M. catarrhalis* results in the underestimation of *M. catarrhalis* in cultures of human respiratory tract samples.

M. catarrhalis produces oxidase, catalase, and DNase. Several kits to speciate *M. catarrhalis* are commercially available.[10,11]

Epidemiology and Respiratory Tract Colonization

M. catarrhalis has been recovered exclusively from humans. The prevalence of colonization is highly dependent on age. The upper respiratory tract of approximately 1% to 5% of healthy adults is colonized by *M. catarrhalis*.[12,13] By contrast, nasopharyngeal colonization with *M. catarrhalis* is common throughout infancy. Some published studies have shown a higher rate of colonization during winter months; this higher rate may be a result of the appearance of respiratory viral illnesses during colder months. Substantial regional differences in colonization rates are observed. For example, 66% of infants in a study in Buffalo, New York, were colonized during the first year of life,[14] whereas a similar study in Göteborg, Sweden, showed a colonization rate of approximately half of that level.[15] A study of rural Aboriginal infants near Darwin, Australia revealed that 100% of infants were colonized by *M. catarrhalis* by the age of 3 months.[16] The explanation for the marked differences in rates of colonization is not yet known. Several factors, including living conditions, daycare, crowding, hygiene, environmental factors (e.g., household smoking), genetic characteristics of the populations, and host factors, may play a role.[17,18] Microbial interactions in the nasopharynx among cocolonizing respiratory pathogens, including viruses and bacteria, and normal flora contribute to the complexity and dynamics of nasopharyngeal colonization.[19–25] The widespread use of pneumococcal conjugate vaccines has caused changes in patterns of nasopharyngeal colonization by reducing colonization with vaccine serotypes of *Streptococcus pneumoniae*. This effect has resulted in "replacement" of vaccine serotypes of *S. pneumoniae* by nonvaccine pneumococcal serotypes. Whether replacement by nontypeable *Haemophilus influenzae* and *M. catarrhalis* also occurs will require careful monitoring.[26–28]

Nasopharyngeal colonization with middle ear pathogens, including *M. catarrhalis*, is associated with otitis media. Early colonization is a risk factor for recurrent otitis media.[14,29,30] Otitis-prone children are colonized with *M. catarrhalis* at a higher rate compared with healthy children.[14,31,32]

M. catarrhalis is isolated from the sputum of adults with chronic obstructive pulmonary disease (COPD). Approximately 50% of episodes of acquisition of *M. catarrhalis* are associated with acute exacerbations of COPD.[33] Adults with COPD develop systemic and mucosal immune responses and clear *M. catarrhalis* from the respiratory tract efficiently after a relatively short duration (<1 month).[34,35] A smaller proportion of episodes of colonization may persist for up to 2 years.[33] Acquisition and clearance of the organism results in strain-specific protection.

Pathogenesis

M. catarrhalis causes mucosal infections in children and adults. The pathogenesis of infection appears to involve contiguous spread of the bacterium from its colonizing position in the respiratory tract to cause clinical signs of infection. In the case of otitis media the isolates recovered from the middle ear are present in the nasopharynx, indicating that the middle ear isolate came from the nasopharynx via the eustachian tube. Colonization of the upper respiratory tract with middle ear pathogens, including *M. catarrhalis*, is a necessary first step in the pathogenesis of otitis media. However, colonization alone is not sufficient to cause disease. An inciting event, such as a viral infection, in a child colonized with a middle ear pathogen is probably necessary for bacteria to move to the middle ear and cause otitis media. In the case of infection in adults with COPD the acquisition of a new strain is critical in the pathogenesis of infection.[33,36]

A key step in the initiation of infection is adherence of *M. catarrhalis* to the human respiratory epithelium. Several adhesins with varying specificities for host cells have been identified (Table 213.1).[3,7,37] Some of these surface antigens and others are being evaluated as potential vaccine antigens.[6,38–44] In addition to adherence to the epithelial surface, *M. catarrhalis* also resides within and beneath the epithelium and invades host cells.[45,46] Indeed, colonization of the nasopharynx by *M. catarrhalis* is more frequent than is revealed by surface culture.

TABLE 213.1 Adhesins and Putative Adhesins of *Moraxella catarrhalis*

ADHESIN	MOLECULAR MASS (kDa)	OBSERVATION
UspA1	88 (oligomer)	Adhesin for respiratory epithelial cells; binds laminin
MID/Hag	200	Hemagglutinin; binds IgD
OMP CD	45	Adhesin for respiratory epithelial cells; binds mucin; OMP A–like protein
McmA	110	Metallopeptidase-like adhesin
MchA1 (MhaB1)	184	Homology with filamentous hemagglutinin of *Bordetella pertussis*
MchA2 (MhaB2)	201	
McaP	66	Adhesin and phospholipase B
OlpA	24	Putative adhesin based on homology with Opa proteins
OMP J	19 16	Exists in two forms; putative adhesins based on homology with Opa proteins
Type 4 pili	16	Also essential for transformation; involved in biofilm formation

Hag, Hemagglutinin; *IgD,* immunoglobulin D; *McaP, M. catarrhalis* adherence protein; *MchA, M. catarrhalis* hemagglutinin-like protein A; *McmA, M. catarrhalis* metallopeptidase-like adhesin A; *MhaB2, M. catarrhalis* (filamentous) hemagglutinin adhesin-like protein B2; *MID, Moraxella* IgD-binding protein; *OMP,* outer membrane protein; *Opa,* opacity associated protein; *Olp,* Opa-like protein; *UspA1,* ubiquitous surface protein A1.

The outer membrane of *M. catarrhalis* contains lipooligosaccharide (LOS). LOS consists of a lipid A core coupled to oligosaccharides. The structure of the LOS resembles that of other nonenteric gram-negative bacteria in that the molecule lacks the long polysaccharide side chains observed in enteric gram-negative bacteria. Three major antigenic types of LOS can be distinguished, accounting for 95% of all strains.[47] The different serotypes are based on differences in terminal sugars in the LOS molecule. A detoxified LOS molecule is a potential vaccine antigen.[48,49]

The interaction of selected surface antigens with receptors of various host cells in the respiratory tract has important effects in mediating host responses to *M. catarrhalis*. For example *M. catarrhalis* is mitogenic for B lymphocytes through interaction with the MID/Hag protein.[50,51] The surface protein UspA2 regulates nuclear factor kappa B and subsequent interleukin-8 release by human respiratory epithelial cells.[52] LOS stimulates human monocytes to produce proinflammatory cytokines in both Toll-like receptor 4– and CD14-dependent pathways.[53] Like many gram-negative bacteria, *M. catarrhalis* sheds vesicles from its surface; these vesicles are internalized by respiratory epithelial cells and mediate several virulence mechanisms, including B-cell activation, induction of inflammation, and delivery of β-lactamase, which may promote survival of copathogens.[54–56] The availability of the genome sequence of clinical isolates is facilitating studies of pathogenesis.[57,58] *M. catarrhalis* evades host immunity through specific binding of several human extracellular matrix molecules in connective tissue, including vitronectin, fibronectin, collagen, plasminogen, and cartilage oligomeric matrix protein.[58–64] These host molecular patterns become exposed when epithelial integrity is interrupted, for example, by chronic inflammation or viral infection.

A reliable animal model that parallels human infection has not yet been developed for *M. catarrhalis*. The specificity of *M. catarrhalis* for humans creates challenges for the development of a useful model to study pathogenesis. The chinchilla model of otitis media is used widely to study otitis media caused by other bacteria, but chinchillas readily clear *M. catarrhalis* when it is instilled into the middle ear. *M. catarrhalis* colonizes the nasopharynx of the chinchilla, so this model may prove useful.[65,66] The most widely used model is a mouse pulmonary clearance model that measures the rate of clearance of *M. catarrhalis* from the lungs after intratracheal challenge. This model does not parallel human infection but has been used as a guide to identify and study potential

vaccine antigens. Models that involve coinfection of *M. catarrhalis* with other viral and respiratory tract pathogens may prove to be useful in understanding the role of *M. catarrhalis* as a copathogen.[67–69]

Clinical Manifestations

Otitis Media

Approximately 80% of children experience at least one episode of acute otitis media by the age of 3 years. A subset of children experiences recurrent otitis media, which is associated with a delay in speech and language development. Careful studies from many centers have defined the cause of acute otitis media by culturing middle ear fluid obtained by tympanocentesis, which has been considered the gold standard for determining the etiology of otitis media. Although some differences among studies are observed, the results from centers in the United States and Europe are remarkably consistent in showing that *S. pneumoniae*, nontypeable *H. influenzae*, and *M. catarrhalis* are the predominant bacterial causes of acute otitis media. Overall, based on cultures of middle ear fluid, approximately 5% to 20% of cases of acute otitis media are caused by *M. catarrhalis*.[70–78] However, culture detects pathogens in only a subset of otitis media.[79] Using more sensitive molecular analysis, *M. catarrhalis* is detected alone or with other pathogens in 30% to 50% of middle ear fluid samples from children with otitis media (Fig. 213.1).[80–86] Analysis of middle ear fluid by PCR increases the frequency of detection of *S. pneumoniae* and nontypeable *H. influenzae* 3.2-fold compared with culture, whereas *M. catarrhalis* is 4.5 times more likely to be identified by PCR.[87] Several lines of evidence indicate that the presence of bacterial DNA in clinical samples indicates active infection, including (1) detection of messenger RNA, (2) viable bacteria in biofilms, and (3) studies in the chinchilla model.[88–90] When *M. catarrhalis* is isolated from middle ear fluid in acute otitis media, it is present as a sole pathogen in approximately 75% of cases and as a copathogen in the remaining 25%. The widespread use of pneumococcal conjugate vaccines is altering the distribution of pathogens in otitis media, emphasizing the importance of continuous monitoring.[91,92]

Lower Respiratory Tract Infections in Chronic Obstructive Pulmonary Disease

M. catarrhalis causes lower respiratory tract infections in adults, particularly in the setting of COPD. The recognition of *M. catarrhalis* as a pathogen in this setting was delayed until the past 20 years because *M. catarrhalis* is indistinguishable from commensal *Neisseria* by Gram stain and difficult to distinguish by colony morphology. Therefore, unless clinical microbiology laboratories specifically test colonies that appear to be *Neisseria, M. catarrhalis* will be missed as a potential pathogen in sputum.

Several lines of evidence have established that *M. catarrhalis* causes exacerbations of COPD[93,94]:

- Analysis of sputum samples of a subset of patients with exacerbations of COPD demonstrates a predominance of gram-negative diplococci on Gram stain and almost pure cultures of *M. catarrhalis.*
- Studies using transtracheal aspiration and bronchoscopy with the protected specimen brush to sample the lower airways have revealed pure cultures of *M. catarrhalis* in some patients with exacerbations of COPD.
- The levels of inflammatory markers in the sputum of patients with exacerbations with positive cultures for *M. catarrhalis* are higher than the level in sputum of culture-negative exacerbations.[95]
- A specific immune response has been observed after exacerbations of COPD associated with *M. catarrhalis* in the sputum.[33–35]
- Acquisition of a new strain of *M. catarrhalis* is associated with clinical exacerbation.[33,36]
- Exacerbations resulting from acquisition of a new strain of *M. catarrhalis* are associated with protease-antiprotease imbalance in the airways.[96,97]

M. catarrhalis causes approximately 10% of exacerbations of COPD, making the bacterium the second most common bacterial cause of exacerbations after nontypeable *H. influenzae*.[3,33,98]

The clinical manifestations of exacerbations of COPD caused by *M. catarrhalis* are similar to those of exacerbations caused by other bacteria, such as nontypeable *H. influenzae.* Patients experience increased cough and sputum production, increased sputum purulence, and increased dyspnea compared with baseline symptoms. Sputum Gram staining shows intracellular and extracellular gram-negative diplococci as the exclusive or predominant bacterial form (Fig. 213.2), and cultures grow predominantly *M. catarrhalis.*

Pneumonia in Older Adults

M. catarrhalis causes pneumonia in older adults, but it is difficult to state the precise proportion. Most older patients who experience pneumonia caused by *M. catarrhalis* have underlying illnesses, including COPD, congestive heart failure, and diabetes. Although *M. catarrhalis* causes a significant illness in older adults, fulminant pneumonia is uncommon.

Nosocomial Respiratory Tract Infections

Nosocomial lower respiratory tract infections caused by *M. catarrhalis* have been observed in respiratory units in health care facilities. The

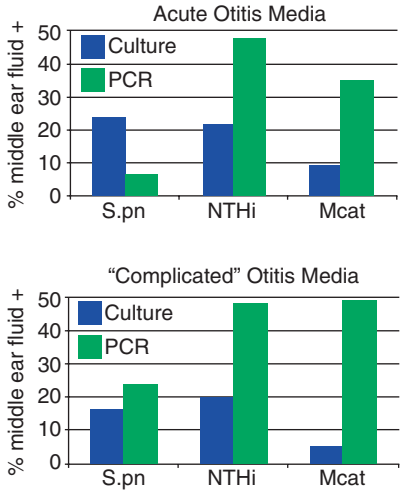

FIG. 213.1 Causes of otitis media. These are the results of bacterial culture *(blue)* and polymerase chain reaction *(PCR) (green)* of middle ear fluid from children with acute otitis media *(top)* and complicated otitis media defined as recurrent, refractory, or chronic otitis media and otitis media with effusion *(bottom).* Studies included 1565 (acute) and 894 (complicated) samples from seven studies.[80–86] *Mcat, Moraxella catarrhalis; NTHi,* Nontypeable *Haemophilus influenzae; S.pn, Streptococcus pneumoniae.*

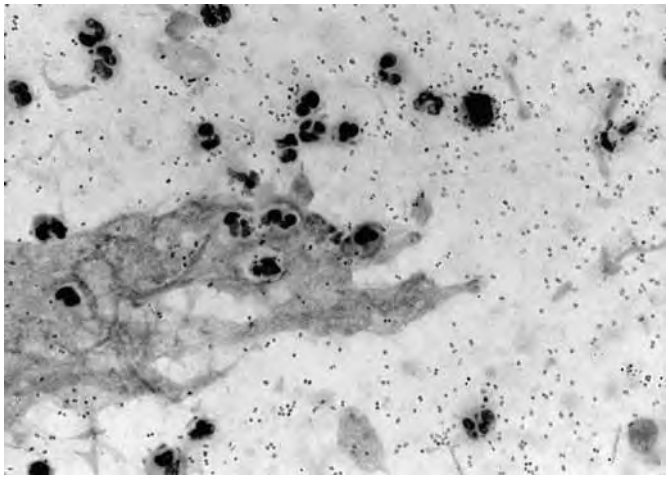

FIG. 213.2 Sputum sample from a patient with chronic obstructive pulmonary disease experiencing an exacerbation caused by *Moraxella catarrhalis*. Note the abundance of leukocytes, the presence of large numbers of gram-negative diplococci as the exclusive bacterial form and the presence of intracellular bacteria in leukocytes (Gram stain; ×1000).

presence of a susceptible population of adults with underlying cardiopulmonary disease may be important in these apparent outbreaks. Analysis of isolates by various typing methods has indicated that some of these clusters involved multiple strains of *M. catarrhalis,* and some were caused by a single strain, indicating person-to-person spread of the organism.[99]

Sinusitis

The cause of sinusitis is determined by culture of sinus aspirates, a relatively invasive procedure that is not performed routinely. Studies that have used sinus aspiration to determine the cause of sinusitis have shown that *M. catarrhalis* is the third most common cause of sinusitis in adults and children after nontypeable *H. influenzae* and *S. pneumoniae.*

Bacteremia

Published reports have documented the occurrence of bacteremia caused by *M. catarrhalis.*[100–102] Bacteremia is an infrequent manifestation of *M. catarrhalis* infection. The severity of clinical manifestations ranges from mild to life threatening. Bacteremia has been reported in those of all ages, from neonates to older adults. Most patients have clinical evidence of respiratory tract infection. Whereas many infants with bacteremia caused by *M. catarrhalis* are immunocompetent, most adults have underlying illnesses, including cardiopulmonary disease, malignancy, immunodeficiency, and chronic debilitation. A review of *M. catarrhalis* bacteremia has noted a mortality of 21%.[102] The underlying illness is an important determinant of outcome.

Therapy

A rapid increase in the proportion of strains that produce β-lactamase occurred simultaneously in the United States and Europe, beginning in the late 1970s.[103] This is one of the most dramatic examples of a rapid increase in antimicrobial resistance by a bacterial species. Currently, almost all strains of *M. catarrhalis* produce β-lactamase. Three different β-lactamases (BRO-1, BRO-2, and BRO-3) have been identified and characterized.[**104**,105] The β-lactamase of *M. catarrhalis* is inducible and cell associated. Because an inoculum-dependent susceptibility to ampicillin is observed, ampicillin should not be used for β-lactamase–producing strains, regardless of the results of susceptibility testing. Antimicrobial susceptibility patterns for *M. catarrhalis* have remained relatively stable. However, the detection of macrolide- and tetracycline-resistant isolates, thus far confined to China and the Asia Pacific region, demands continued vigilance.[**106**–111]

Most infections caused by *M. catarrhalis* can be treated with oral antibiotics. The organism is generally susceptible to amoxicillin-clavulanate, trimethoprim-sulfamethoxazole, tetracyclines, oral cephalosporins (e.g., cefixime, cefpodoxime, cefprozil, cefaclor, cefuroxime, and others), macrolides (e.g., azithromycin, clarithromycin), and fluoroquinolones. *M. catarrhalis* is also uniformly susceptible to ticarcillin, piperacillin, second- and third-generation cephalosporins, and aminoglycosides. *M. catarrhalis* is resistant to penicillin, ampicillin, vancomycin, and clindamycin.

Vaccines to prevent *M. catarrhalis* infections are not yet available, but several candidate vaccine antigens are under active investigation.[**112**–**114**]

OTHER *NEISSERIA*

N. meningitidis and *N. gonorrhoeae* have long been recognized as the pathogenic *Neisseria.* Other *Neisseria* spp. are common components of the normal flora of the upper respiratory tract of humans and are often called commensal *Neisseria.* Table 213.2 lists several biochemical and growth characteristics used to distinguish among various *Neisseria* spp. and *M. catarrhalis.* Commensal *Neisseria* spp. lack several virulence factors, including pili, opacity-associated (Opa) proteins, and the H8 antigen, that are expressed by meningococci and gonococci and have an altered lipid A structure compared with that in the LOS of pathogenic *Neisseria.*[115,**116**] However, commensal *Neisseria* express a variety of surface antigens that share homology with antigens of *N. meningitidis* and *N. gonorrhoeae.*[117,118] Immune responses to cross-reactive antigens on commensal *Neisseria*, particularly *N. lactamica*, may contribute to acquisition of natural immunity to *N. meningitidis,* and this characteristic is under evaluation as a potential approach to vaccine development.[119–121]

Neisseria spp. such as *N. sicca, N. subflava, N. cinerea, N. lactamica*, and others occasionally cause invasive infections in humans. These infections, documented primarily by individual case reports, include meningitis, endocarditis, bacteremia, ocular infections, pericarditis, empyema, peritonitis, septic arthritis, bursitis, and osteomyelitis. *N. cinerea* appears to have a propensity to cause ocular infections in young children.

N. weaveri (formerly Centers for Disease Control and Prevention group M5) is a component of the normal oropharyngeal flora of dogs and is an important cause of infection in dog bite wounds in humans. These infections are occasionally associated with bacteremia.

Many of these infections have been treated successfully with penicillin and ampicillin. However, isolates of *Neisseria* spp. have shown increased resistance to penicillin, so susceptibility testing should be performed on isolates that cause invasive infections, and the results should be used to guide antimicrobial therapy.

Neisseria are naturally competent for uptake of DNA. Genetic recombination occurs among bacteria that make up the complex flora of the upper respiratory tract. Virulence determinants are exchanged with pathogenic *Neisseria*, genes encoding altered penicillin-binding proteins are passed between species, and extensive interspecies recombination of a variety of genes occurs in vivo.[122–126] These observations have important implications in the role of acquisition of antibiotic resistance and evolution of pathogens in the human respiratory tract. Indeed, interspecies transfer of *penA* genes from commensal *Neisseria* is an important mechanism of acquisition of penicillin resistance of *N. gonorrhoeae* and *N. meningitidis.*[122]

OTHER *MORAXELLA*

Bacteria of the genus *Moraxella* are normal commensals of the human upper respiratory tract and are occasionally recovered from the skin and urogenital tract. Species are generally differentiated biochemically. Table 213.3 shows biochemical reactions and growth characteristics used to distinguish among several *Moraxella* spp.

Moraxella spp. other than *M. catarrhalis* are unusual pathogens in humans. Several reports have emphasized the role of other *Moraxella* as ocular pathogens. These bacteria cause conjunctivitis, keratitis and, rarely, endophthalmitis.[127–129] Most episodes of *Moraxella* keratitis have predisposing local ocular pathology. Healing may take weeks to months. *Moraxella* spp. are susceptible to all conventional topical ocular antibiotics.

Case reports have established *Moraxella* spp. as unusual causes of invasive infections in humans, including endocarditis, bacteremia, septic arthritis, purulent pericarditis, cellulitis, and meningitis. Patients who experience meningitis caused by *Moraxella* spp. have a high frequency of inherited and acquired complement deficiencies, and these should be investigated after recovery.[130]

Antimicrobial susceptibility should be performed on isolates recovered from normally sterile sites, but *Moraxella* spp. are generally susceptible to penicillins and cephalosporins.

KINGELLA

History and Microbiology

Current taxonomic classification schemes place *Kingella* in the family Neisseriaceae. *Kingella* are short gram-negative cocci to medium-sized rods with tapered ends. Four species have been identified: *K. kingae, K. indologenes, K. denitrificans*, and *K. oralis. Kingella* spp. are recovered from the human respiratory tract and have previously been recognized as rare causes of human disease. However, in the past 2 decades infections caused by *Kingella* have been recognized with increasing frequency.[131]

The increase in the recognition of *Kingella* infections is a result of several factors. Because the bacterium is slow growing and fastidious, special attention by the microbiology laboratory is often required to isolate the organism. Another factor is that the bacterium has likely been misidentified as *Moraxella* and other *Neisseria* by many laboratories. Finally, the development and application of PCR-based diagnostic testing, along with an elevated index of suspicion, has been important in recognizing this emerging pathogen in children.[131–135] *K. kingae*, the most common human pathogen of the *Kingella* spp., grows on blood and chocolate agar but fails to grow on MacConkey agar. The bacterium has a tendency

TABLE 213.2 Biochemical and Growth Characteristics of *Neisseria* and *Moraxella catarrhalis*

	PRODUCTION OF ACID FROM										
SPECIES	**GLUCOSE**	**MALTOSE**	**SUCROSE**	**LACTOSE (ONPG)**	**FRUCTOSE**	**H_2S[a]**	**OXIDASE**	**EXTRA CO_2**	**GROWTH AT 22°C**	**POLYSACCHARIDE[b]**	**PIGMENT[c]**
N. gonorrhoeae	+	—	—	—	—	—	+	VI	—	NG	—
N. meningitidis	+	+	—	—	—	—	+	I	—	NG	—
N. lactamica	+	+	—	+	—	+	+	v	v	—	+ Y
N. sicca	+	+	+	—	+	+	+	—	+	+	— (slY)
N. subflava	+	+	v	—	v	+	+	—	+	v	+ Y
N. mucosa	+	+	+	—	+	+	+	—	+	+	– (slY)
N. flavescens	—	—	—	—	—	+	+	—	+	+	+ Y
N. cinerea	—	—	—	—	—		+	—	v	—	Grayish
N. polysaccharea	+	+	—	—	—		+		—	+	— (slY)
N. elongata	—	—	—	—	—		+		+	—	Grayish/slY
N. weaveri	—	—	—	—	—	+	+		+	—	— (slY)
M. catarrhalis	—	—	+	—	—	+	+	—	v	—	Grayish

[a]With lead acetate paper.
[b]Synthesis of polysaccharide from 5% sucrose.
[c]On Loeffler slant.
CO_2, Carbon dioxide; *H_2S*, hydrogen sulfide; *I*, important for growth; *NG*, no growth; *ONPG*, *o*-nitrophenol-β-D-galactopyranoside; *sl*, slightly; *VI*, very important for growth; *v*, variable; *Y*, yellow.
From Gröschel DM. Moraxella catarrhalis and other gram-negative cocci. In: Mandell GL, Bennett JE, Dolin R, eds. Principles and Practice of Infectious Diseases. *4th ed. New York: Churchill Livingstone; 1995:1926.*

TABLE 213.3 Laboratory Procedures Useful for Identification of *Moraxella, Oligella, Moraxella*-like Organisms, and *Kingella*

SPECIES	MOTILITY	OXIDASE	CATALASE	OXIDATION/ FERMENTATION GLUCOSE	SERUM REQUIRED	UREASE	INDOLE	NITRATE	PHENYLALANINE	GELATIN	ASSIMILATION OF ACETATE	GROWTH ON MACCONKEY AGAR
M. catarrhalis	—	+	+	—	—	—	—	—	—	—	v	v
M. lacunata	—	+	+	—	+	—	—	+	v	v	—	—
M. nonliquefaciens	—	+	+	—	v	—	—	+	—	—	—	—
M. osloensis	—	+	+	—	—	—	—	v	v	—	+	v
M. phenylpyruvica	—	+	+	—	—	+	—	v	+	—	v	v
M. atlantae	—	+	+	—	+	—	—	—	—	—	v	+
O. urethralis	—	+	+	—	—	—	—	—	+	—	v	+
Neisseria weaveri (M5)	—	+	+	—	—	—	—	—	+	—	—	—
N. elongata (M6)	—	+	—	—	—	—	—	+	—	—	v	v
K. kingae[a]	—	+	—	F[b]	—	—	—	—	—	—		v
K. indologenes	—	+	—	F	—	—	+	—	—	—		
K. denitrificans	—	+	—	F	—	—	—	+	—	—		—

[a]Most strains hemolytic on blood agar.
[b]May take 3 or more days; some strains require serum supplement.
F, Fermentation; *v,* variable.
From Gröschel DM. Moraxella catarrhalis and other gram-negative cocci. In: Mandell GL, Bennett JE, Dolin R, eds. Principles and Practice of Infectious Diseases. *4th ed. New York: Churchill Livingstone; 1995:1926.*

to resist decolorization and may therefore sometimes be mistaken for a gram-positive organism. It is oxidase positive, produces acid from glucose and maltose, and lacks catalase, urease, and indole. Table 213.3 lists several characteristics that distinguish *Kingella* from related bacteria. Matrix-assisted laser desorption/ionization time-of-flight mass spectrometry and 16S ribosomal DNA sequencing are also accurate means of identifying *Kingella* spp.

With the recognition of *K. kingae* as a human pathogen, recent work is beginning to shed light on the genetic population structure of the species and on mechanisms of pathogenesis. Genotyping of clinical isolates suggests the presence of substantial genetic heterogeneity with a subset of clones that appear to be associated with invasive disease.[136–139] Key virulence factors include type IV pili, a polysaccharide capsule, and a "repeat-in-toxin" toxin with wide cellular specificity has been identified in *K. kingae*.[140–146]

Epidemiology and Respiratory Tract Colonization

K. kingae frequently colonizes the throats of young children. Blood agar medium with vancomycin 2 μg/mL is used to improve isolation from the oropharynx. The organism has not been recovered from cultures of the nasopharynx. The highest rate of colonization is observed in children ages 6 months to 4 years, which corresponds to the peak age incidence of invasive disease. Infants younger than 6 months are not colonized. This pattern of colonization parallels that of other respiratory tract pathogens, such as *M. catarrhalis* and *S. pneumoniae*, which show low rates of colonization in the neonatal period (presumably as a result of maternal antibodies), followed by higher rates of colonization and infection in infancy and childhood, with a declining incidence of infection in adulthood. *K. kingae* is transmitted person to person, and outbreaks of invasive infection have been observed in daycare centers.[139,142–153]

In studies of the microbiology of the human oral cavity a bacterium that resembled *Eikenella corrodens* was recovered frequently from dental plaque.[154] On the basis of 16S ribosomal sequences, these organisms were designated as the new species *K. oralis*.[155] *K. oralis* is present in plaque or on the tooth surface in most people with or without periodontal disease.[156] The role of *K. oralis* in periodontal disease is not known at this time.

Clinical Manifestations

K. kingae is the most frequent human pathogen of the *Kingella* spp. Approximately 90% of invasive disease caused by *K. kingae* occurs in previously healthy children younger than 4 years. Underlying medical conditions are generally present in older children who experience infections caused by *K. kingae*.[157] Invasive infections have not been reported in infants younger than 6 months. Infection shows a seasonal distribution, with the rate of cases being higher in the autumn and winter months.[158] Outbreaks in daycare centers and closed communities are well documented.[143] The most common clinical manifestations of *K. kingae* disease are septic arthritis, osteomyelitis, tenosynovitis, spondylodiskitis, endocarditis, and bacteremia.[159] The clinical presentation of *K. kingae* infections is often subtle, with low-grade fever and normal levels of acute-phase reactants, emphasizing the importance of a high index of suspicion.[131,160–162]

Skeletal Infections

K. kingae shows a propensity to cause osteoarticular infections in young children and is now recognized as a common cause of such infections.[132,163–167] The disease most frequently presents as septic arthritis of large weight-bearing joints, especially the knee and ankle. Gram staining of the joint fluid is usually negative. The diagnosis is made by recovering the organism from culture of joint fluid. Inoculating blood culture bottles with joint fluid substantially enhances the likelihood of recovering the organism, compared with direct inoculation of agar plates, and should be done routinely with joint fluid samples from young children.[168,169] PCR-based assays have revealed that *K. kingae* is a more common cause of osteoarticular infection than is revealed by culture.[131,132,150,170,171] Osteomyelitis caused by *K. kingae* most frequently involves the bones of the lower extremity. The onset is insidious, and the diagnosis is often delayed. Hematogenous invasion of the intervertebral disk by *K. kingae* is observed most commonly in the lumbar intervertebral spaces but can occur at any level.[150,166,172]

Endocarditis

In contrast to other clinical manifestations of *Kingella* infections, endocarditis can be seen at all ages, including school-age children and adults. Endocarditis has involved native and prosthetic valves. Although many cases of endocarditis occur in those who have preexisting valvular disease, *Kingella* can cause endocarditis on normal valves as well. Cases of endocarditis can be caused by *K. denitrificans* and *K. indologenes* as well as *K. kingae*. *Kingella* spp. are part of the so-called HACEK group of organisms, which include fastidious bacteria capable of causing endocarditis. The HACEK group consists of the following: *Haemophilus parainfluenzae*; three *Aggregatibacter* spp.—*A. (Haemophilus) aphrophilus, A. (Haemophilus) paraphrophilus*, and *A. (Actinobacillus) actinomycetemcomitans; Cardiobacterium hominis; Eikenella corrodens*; and *K. kingae*. The difficulty in recovering and identifying *K. kingae* in blood cultures frequently results in a delay in the diagnosis, which may account for the relatively high rate of morbidity seen with *Kingella* endocarditis. Because of the serious nature of *Kingella* endocarditis, all patients with *Kingella* bacteremia should be carefully evaluated for the presence of endocarditis.

Bacteremia

Approximately 50% of children with *K. kingae* bacteremia have a concomitant focal source, such as the skeletal system. The remainder have occult bacteremia. The presumed source of the bacteremia is the respiratory tract.

Other Infections

Kingella spp. have been documented by case reports to cause infections in a variety of sites, including pneumonia, epiglottitis, meningitis, soft tissue infections, and ocular infections.

Therapy

Kingella spp. are susceptible to a wide variety of penicillins and cephalosporins, although rare isolates of β-lactamase–producing *K. kingae* have been identified.[173–175] Disease-associated isolates should be tested for antimicrobial susceptibility; if the isolate is susceptible, a penicillin or cephalosporin should be used. Other agents with in vitro activity include aminoglycosides, trimethoprim-sulfamethoxazole, tetracycline, erythromycin, and fluoroquinolones. Although specific guidelines for treatment are not yet established, a rational approach is to administer antibiotic therapy for 2 to 3 weeks for *K. kingae* septic arthritis, for 3 to 6 weeks for osteomyelitis, and 3 to 12 weeks for spondylodiskitis. Children are often treated initially with a β-lactam antibiotic intravenously, followed by oral administration once the clinical condition has improved.[131] Attempts to stop outbreaks in daycare centers with prophylactic antimicrobial drugs, such as rifampin, have met with mixed success.[143]

The drugs of choice for the treatment of endocarditis caused by *Kingella* spp. (and other HACEK organisms) are the third-generation cephalosporins cefotaxime or ceftriaxone.[176] The duration of treatment for native valve endocarditis should be 3 to 4 weeks, and the duration of treatment for prosthetic valve endocarditis should be 6 weeks.

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The complete reference list is available online at Expert Consult.

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214 *Vibrio cholerae*

Matthew K. Waldor and Edward T. Ryan

SHORT VIEW SUMMARY

Microbiology and Epidemiology

- *Vibrio cholerae* is a curved motile gram-negative bacillus.
- *V. cholerae* is a noninvasive intestinal pathogen.
- *V. cholerae* O1 and O139 serogroup organisms are the causes of epidemic cholera.
- Non-O1 and non-O139 *V. cholerae* can cause isolated cases of usually mild gastroenteritis.
- Cholera results from secretory diarrhea caused by the actions of cholera toxin (CT) in the intestine.
- CT is an adenosine diphosphate–ribosylating enzyme that leads to chloride, sodium, and water loss from intestinal epithelial cells.
- *V. cholerae* has an aquatic reservoir, particularly in brackish estuarine water.
- Fecal-oral transmission is associated with unsafe water and inadequate sanitation.
- There are an estimated 2 to 3 million cholera cases, resulting in approximately 100,000 deaths each year.
- The current cholera global pandemic began in 1961 and represents the seventh in the historical record.
- Cholera is now endemic in more than 50 countries.
- Cholera can lead to explosive epidemics and outbreaks.
- The current pandemic is caused by *V. cholerae* O1 El Tor organisms, with the largest burdens in South Asia, sub-Saharan Africa, Haiti, and Yemen.

Clinical Manifestations

- Acute severe watery diarrhea can result in death from dehydration within 6 to 12 hours of onset of clinical symptoms.
- Bowel movements during cholera can become progressively more watery, eventually resembling rice water with a fishy odor.
- Vomiting, ileus, and muscle cramps are common.
- The presence of fever should prompt consideration of additional diagnoses.
- Complications of cholera largely are those resulting from hypotension and hypoperfusion and may include acute renal tubular necrosis and stroke, usually in older individuals. Aspiration pneumonia from vomiting may also occur. Death from cholera almost always results from dehydration.

Diagnosis

- Cholera should be considered when an adult or child 5 years of age or older develops severe dehydration or dies of acute watery diarrhea in any area or when an individual 2 years of age or older develops acute watery diarrhea in an area known to be endemic for cholera.
- Rapid antigen tests are available, including dipstick stool assays.
- Confirmatory microbiologic culturing permits definitive identification and assessment of antimicrobial resistance profiles.

Management

- The cornerstone of management is rapid assessment of the degree of dehydration, followed by prompt fluid restoration (see Table 214.2) and vigorous monitoring and matching of ongoing fluid losses.
- Severe dehydration (>10% fluid loss) may require greater than 100 mL/kg of fluid replacement to restore euvolemia, ideally administered within the first 3 to 4 hours of clinical presentation (in 6 hours for children younger than 1 year).
- Additional stool losses should be matched at approximately 10 to 20 mL/kg per diarrheal stool or vomiting episode.
- Moderate dehydration (5%–10% fluid loss) may require 75 to 100 mL/kg of fluid resuscitation within the first 3 to 4 hours of clinical presentation. Patients with no or mild dehydration may be treated with oral rehydration solution (ORS) alone or liquid ad libitum.
- Intravenous fluids are indicated in patients with severe dehydration and in patients unable to ingest adequate ORS. Lactated Ringer solution supplemented with 5% dextrose (ideally supplemented with additional potassium) is an optimal choice for patients with cholera, although intravenous normal saline or normal saline supplemented with dextrose (and potassium) can also be used.
- Oral rehydration treatment should be encouraged in all patients able to ingest, regardless of degree of dehydration and regardless of potential ongoing administration of intravenous fluid.
- ORS can be made in resource-limited settings by adding ½ tsp of table salt to 6 tsp of table sugar in 1 L of safe water, ideally supplemented with locally available potassium sources such as coconut milk, bananas, or orange juice.
- Antibiotics play a secondary role in treatment of patients with cholera; usually a macrolide or fluoroquinolone antibiotic is administered. Tetracyclines can be used in nonpregnant patients older than 7 years in areas with confirmed susceptibility (see Table 214.3).
- Antibiotics decrease the duration of diarrhea and may limit secondary transmission.
- Infants younger than 6 months should also receive 10 mg of zinc each day for 10 days; children 6 months to 5 years of age should receive 20 mg of zinc each day for 10 days.

Prevention

- Control primarily focuses on surveillance, case detection, fluid resuscitation and management, vaccination, and provision of safe water and adequate sanitation.
- A number of oral killed cholera vaccines are currently commercially available and approved by the World Health Organization for global use (see Table 214.4).
- A live attenuated oral cholera vaccine is approved by the US Food and Drug Administration.
- Additional cholera vaccines are under development.

Cholera is an acute and often severe watery diarrheal disease that is among the most rapidly fatal infectious diseases of humans. Previously well individuals infected with *Vibrio cholerae,* the comma-shaped gram-negative rod that causes this disease, can die from dehydration in less than a day. Two epidemiologic features of cholera also distinguish it from other diarrheal diseases. First, cholera can appear in explosive epidemics, particularly among populations that lack access to clean water and adequate sanitation and who are immunologically naïve to the organism. Second, cholera occurs in pandemics, where one strain of *V. cholerae* spreads around the world in essentially clonal fashion. The ongoing seventh pandemic of cholera began in 1961 on the Indonesian island of Sulawesi. Cholera is thought to have afflicted human populations for centuries, if not longer, particularly on the Indian subcontinent.[1] Throughout the centuries, cholera epidemics have influenced human history and killed millions. Studies of cholera and *V. cholerae* have had a broad impact in several scientific fields, and rehydration strategies developed for the treatment of cholera have been extremely useful for treatment of other diarrheal diseases.[2] Snow's studies in London in the 1840–50s linking the spread of cholera to the water supply are considered central to the establishment of the field of epidemiology. Koch isolated *V. cholerae* from patients with cholera in Calcutta (Kolkata) in 1883; however, Pacini likely detected the organism in 1854 in the intestines of cholera victims in Florence.[3]

CLASSIFICATION AND GENOMICS

V. cholerae organisms are oxidase-positive facultative anaerobes that can be identified using a variety of biochemical tests and selective media.[4] The organisms are characteristically highly motile and bear a unipolar sheathed flagellum. *V. cholerae* is classified into more than 200 serogroups; differences in the composition of the O-specific polysaccharide (OSP) chains of lipopolysaccharide (LPS) molecules establish the chemical basis for the distinct antigenicity of the serogroups. The O1 serogroup of *V. cholerae* was the only serogroup associated with epidemic cholera until 1992, when *V. cholerae* O139 emerged. However, some non-O1, non-O139 *V. cholerae* serogroups have given rise to gastroenteritis outbreaks, but not to large epidemics. In general, the majority of *V. cholerae* isolates from the environment including those of *V. cholerae* O1 do not produce the virulence factors, such as cholera toxin (CT), that are required to cause cholera. The O1 serogroup is divided into two principal serotypes, Inaba and Ogawa, which differ by the presence of a 2-*O*-methyl group in the nonreducing terminal sugar of the Ogawa OSP, which is absent from Inaba OSP.[5] Switching of serotypes during epidemics is well described; the driving forces underlying switching are not known, but they do not appear to be random.[6] CT-producing (toxigenic) O1 serogroup strains are also divided into two biotypes based on a number of microbiologic and biochemical differences.[4] The classic biotype of *V. cholerae* O1 caused the second[7] and sixth cholera pandemics and probably the earlier and intervening pandemics as well, whereas the El Tor biotype of *V. cholerae* O1 is the cause of the ongoing seventh cholera pandemic. The classic biotype of *V. cholerae* is now likely extinct.

V. cholerae, similar to many other bacterial enteric pathogens, is classified as a Gammaproteobacteria. In contrast to most Gammaproteobacteria such as *Escherichia coli,* whose genomes consist of single circular chromosomes, the genome of *V. cholerae* is multipartite. The *V. cholerae* genome (and that of all *Vibrio* spp.) is unequally divided between two circular chromosomes.[8] Its large chromosome (≈3 kb) contains the vast majority of the essential genes as well as genes implicated in pathogenicity, whereas the small chromosome (≈1 kb) is enriched for genes of unknown function. A plausible explanation for the evolution of the bipartite *V. cholerae* genome is that a remote ancestor of *V. cholerae* (and all vibrios) acquired a large plasmid that subsequently obtained essential genes and other chromosome-like features.[9] Acquisition of virulence-linked genes from other (unknown) donor organisms via lateral gene transfer has played a central role in the evolution of pathogenic *V. cholerae*.[10] For example, the genes encoding CT are encoded within a bacteriophage that infected and integrated its DNA into the genome of a nontoxigenic precursor of toxigenic *V. cholerae* O1.

PATHOGENICITY

V. cholerae has no known natural vertebrate host besides humans. However, the pathogen is not dependent on humans for its propagation; *V. cholerae* grows in brackish estuaries and coastal seawaters, often in close association with copepods or other zooplankton. *V. cholerae* can also grow in water of lower salinity when it is warm and adequate organic material is available.[11] Because patients with severe cholera can excrete up to 20 L of "rice water" stool laden with ≈10^9 *V. cholerae* cells/mL per day, humans greatly facilitate the propagation and dissemination of the pathogen. Humans usually become infected with *V. cholerae* after ingestion of contaminated water or food, although there may be direct fecal-oral spread between people that does not involve water or food. The infectious dose in volunteer studies carried out in the United States is relatively high.[12] Low stomach pH is known to lower the infectious dose in volunteers; additional factors likely impact the infectious dose as well.[12] Because the frequency of hypochlorhydria (caused by chronic *Helicobacter pylori* infection) in cholera endemic regions is often high, the *V. cholerae* infectious dose may be considerably lower in these regions.

After passage through the stomach, *V. cholerae* has the unusual capacity to survive and multiply in the small intestine. Although many processes and gene products contribute to the organism's ability to colonize the small intestine,[13] toxin coregulated pili (TCP) are the most critical identified factors primarily dedicated to mediating colonization.[14] The major subunit of these pili is encoded by *tcpA,* the first gene of the *tcp* operon. Most of the other genes of the operon encode the machinery for the biogenesis of TCP,[15] but *tcpE* encodes a protein found at the tip of the pilus, and *tcpF* encodes a protein secreted by the TCP assembly apparatus that is also required for colonization.[16] TCP are thought to promote *V. cholerae* intestinal colonization in at least three ways: TCP (1) mediate bacterium-bacterium interactions, enabling microcolony formation within the small intestine; (2) confer protection against toxic factors produced in the intestine; and (3) promote attachment to the intestinal epithelium.[15] The *tcp* operon is found within the TCP pathogenicity island, a chromosome segment ≈41 kb that also encodes additional virulence-associated factors,[17,18] which appears to have been acquired via horizontal gene transfer by a nonpathogenic ancestor of contemporary *V. cholerae*. In addition, the type VI secretion system may be induced in the intestine, promoting colonization potentially by enabling interactions with the microbiota.[19]

While colonizing the small intestine, *V. cholerae* secretes CT, the AB_5 subunit–type protein toxin that causes the secretory diarrhea that is characteristic of cholera. The sufficiency of CT to elicit cholera-like diarrhea was demonstrated experimentally; volunteers fed as little as 5 µg of purified CT developed severe diarrhea indistinguishable from cholera.[20] Thus cholera is a toxin-mediated disease, and *V. cholerae* is a noninvasive mucosal pathogen. The pentameric B subunit of CT binds to GM1, a glycosphingolipid on the surface of epithelial cells. The CT-GM1 interaction targets the toxin through a retrograde trafficking pathway from the plasma membrane to the Golgi apparatus and endoplasmic reticulum and then to the cytoplasm.[21] In the cytoplasm, the enzymatic A subunit of CT mediates the transfer of adenosine diphosphate ribose from nicotinamide adenine dinucleotide to the G protein that regulates adenylate cyclase activity, leading to elevation in the intracellular cyclic adenosine monophosphate (cAMP) concentration.[22] Increased cAMP levels promote chloride ion (Cl^-) secretion by intestinal crypt cells and decreased absorption by villous cells. Water moves from epithelial cells into the bowel lumen to maintain osmolality, and diarrhea results when the resorptive capacity of the remainder of the gut is exceeded. Additional actions of CT (besides dysregulation of adenylate cyclase) likely contribute to the secretory response underlying cholera.[22] Also, other factors besides CT contribute to the diarrheal response because volunteers who ingest *V. cholerae* deficient for *ctxA* often develop mild diarrhea.[23]

The genes encoding CT, *ctxA* and *ctxB (ctxAB)*, are not part of the ancestral *V. cholerae* genome; instead, these genes are embedded in the genome of CTXΦ, a filamentous bacteriophage that infected *V. cholerae,* rendering it toxigenic.[24] Of note, the receptor for this phage is TCP, suggesting that a TCP+, *ctxAB*– strain was the precursor of contemporary pathogenic *V. cholerae*. The intricate interdependence of these two mobile elements—the TCP pathogenicity island and the CTX prophage—extends

even further; ToxT, a key transcriptional activator of *ctxAB* expression, similar to the *tcp* operon, is encoded within the TCP island. Mobile genetic elements such as phages, plasmids, integrative conjugative elements, and pathogenicity islands have played critical roles in the evolution of most bacterial pathogens.

Many of the key virulence factors of *V. cholerae* such as CT and TCP are not constitutively expressed. Instead, fairly shortly after *V. cholerae* is ingested, as yet undefined host signals trigger the coordinated expression of the principal virulence factors of *V. cholerae*.[14] A membrane-embedded transcription activator, ToxR, sits at the top of a signaling cascade that promotes expression of many of the pathogen's virulence-associated genes.[25] There may be a specific temporal order of *V. cholerae* gene expression during expression.[26] Furthermore, as the pathogen passes to more distal parts of the intestine, expression of late genes promotes *V. cholerae* survival outside the human host and engenders a transient hyperinfectious state, which promotes transmission of the organism to new hosts.[27,28] Additional regulatory pathways such as quorum sensing systems, which modify gene expression according to bacterial density and the concentrations of autoinducer molecules, modulate expression of *V. cholerae* virulence. Notably, it has been suggested that crosstalk between the quorum sensing molecules of the microbiota and quorum regulation of *V. cholerae* impact the pathogen's capacity to colonize the intestine.[29]

EPIDEMIOLOGY

Clinical descriptions of cholera-like illness exist in ancient Sanskrit texts, and cholera has likely been endemic to the Ganges delta regions for centuries, if not longer. Seven cholera pandemics have been recorded since 1817. All the pandemics are thought to have originated in Asia, particularly from the Indian subcontinent. The El Tor O1 *V. cholerae* strain causing the ongoing seventh pandemic is evolving. Polymorphisms in the genome of the pandemic strain have been reported to have arisen in the Bay of Bengal and then to have independently spread to other continents.[30] All seven pandemics were likely caused by *V. cholerae* O1, but in 1992 *V. cholerae* O139 emerged in India and Bangladesh to become the first non-O1 serogroup to give rise to epidemic cholera. Although *V. cholerae* O139 initially caused large cholera outbreaks, this serogroup is not a significant cause of disease anywhere at the present time; additional O139 cases were never reported outside of Asia. Horizontal gene exchange appears to explain the molecular origins of *V. cholerae* O139; the genome of this novel *V. cholerae* serogroup is essentially identical to that of El Tor O1 *V. cholerae* except that the genes encoding O1 antigen biosynthesis have been replaced by genes encoding the O139 O-antigen.[31]

Cholera had been absent from the Americas for nearly a century until 1991, when a large outbreak began in Peru and rapidly spread throughout much of South and Central America. During the ensuing decade, more than 1 million cases were reported. Whole-genome–based epidemiologic analyses suggest that there were two separate introductions of El Tor O1 strains, from Africa and Asia, that gave rise to this epidemic.[32] Cholera did not reach the Caribbean during this outbreak. However, in October 2010, a severe cholera epidemic began in Haiti on the Caribbean island of Hispaniola. Since the onset of the epidemic, more than 800,000 cases and 9500 deaths have been recorded in Haiti, and focal related outbreaks and cases have occurred throughout the Caribbean basin.[33] Several lines of evidence indicate that cholera may have been introduced to Haiti by United Nations security forces from Nepal,[34,35,36] suggesting that asymptomatic carriers of *V. cholerae* can play an important role in transmitting cholera over long distances. In October 2016 a large cholera outbreak began in the capital city of Yemen, and by September 2017 almost the entire country was affected, resulting in more than 700,000 cases and 2000 deaths.[37]

The number of cholera cases reported to the World Health Organization (WHO) varies by year depending on the size and extent of outbreaks and endemic disease. In 2011 there were approximately 600,000 cases of cholera and nearly 8000 cholera-related deaths reported to the World Health Organization from 58 countries (Fig. 214.1).[38] In 2015 there were approximately 170,000 cases and 1300 deaths reported from 42 countries.[39] Due to large outbreaks, including in Yemen, these numbers increased dramatically to 1,227,391 cases and 5654 deaths reported from 34 countries in 2017.[39a] These estimates of cholera morbidity and mortality significantly underrepresent the true burden of this disease because many cases are not diagnosed or reported. More recent estimates suggest that approximately 2 to 3 million cholera cases occur annually worldwide with 100,000 deaths.[40] In the decade preceding the outbreaks in Haiti and Yemen, more than 90% of reported cholera cases were from Africa, although the burden of disease in Asia is significantly underreported. More recent analysis suggests that cholera outbreaks in Africa represent repetitive reintroductions of *V. cholerae* from Asia.[41]

CLINICAL MANIFESTATIONS

Cholera manifests as an acute, often severe watery diarrhea that can lead to death from dehydration within hours of onset. The spectrum of disease in individuals infected with *V. cholerae* includes asymptomatic intestinal colonization of *V. cholerae,* observed in individuals with preexisting immunity and mild, moderate, and severe diarrhea. In severe cholera the volume of watery diarrhea can exceed 1 L/h. Vomiting is common. In the initial stages, diarrhea may contain intestinal contents; however, as diarrhea progresses, it becomes more watery, eventually becoming clear, with flecks of white mucus (rice water stool) with a fishy odor. Individuals with cholera are usually afebrile and may be hypothermic because of severe dehydration. The extensive fluid secretion into the intestinal lumen can lead to abdominal cramping and discomfort, and the severe dehydration and perturbations in calcium and potassium levels can lead to ileus, muscle pain and spasm, and tetany. In severely dehydrated patients the skin can become mottled or darkened, reflecting the hyperviscosity and poor circulation of blood.

The clinical manifestations of severe cholera reflect the degree of dehydration. Individuals may be lethargic or unresponsive, have sunken eyes, and have markedly decreased skin turgor often leading to skin tenting (i.e., when skin is pulled back, it takes a long time for it to fall back into place). Individuals may have cold and clammy skin, have dry mucous membranes, and be anuric. Extensive loss of bicarbonate in stool and lactic acidosis secondary to compromised circulation and perfusion can lead to Kussmaul breathing.[42] The peripheral pulse is often rapid in the initial stages of dehydration; however, it can become progressively fainter, eventually becoming nonpalpable as dehydration progresses. Hypoglycemia can be severe, especially in children with limited glycogen stores, and can contribute to lethargy, seizures, and coma. Paradoxically, stool volume can decrease as the degree of dehydration increases, giving a false impression of improvement before total circulatory collapse. Cholera is an acute illness, and the complications of cholera reflect severe hypoperfusion and can include stroke, acute tubular necrosis with renal dysfunction, and aspiration pneumonia from the extensive vomiting.[43] Diarrhea is usually most prominent for 1 to 3 days.

The clinical manifestations of cholera caused by *V. cholerae* O1 and O139 are indistinguishable. Similarly the disease caused by the Ogawa and Inaba serotypes of *V. cholerae* O1 is indistinguishable. At the population level, disease caused by classic biotype *V. cholerae* O1 is more severe than that caused by O1 El Tor biotype strains. Differences in the amino-acid sequences of the CT produced by the two biotypes may account for the heightened severity of diarrhea associated with the classic biotype. So-called variant (or hybrid) strains of *V. cholerae* O1 El Tor that express classic biotype CT may be associated with more severe clinical manifestations than traditional El Tor strains, although the reason for this increased severity is unclear.[44]

Host factors likely also contribute to the clinical manifestations of cholera. For example, when *V. cholerae* is first introduced into an immunologically naïve population (e.g., into Haiti in 2010), individuals of all ages can be equally affected, and disease is often severe. In comparison, in regions where cholera is endemic, many older children and adults have preexisting immunity, and these individuals can be asymptomatically colonized or present with mild-to-moderate diarrhea.[45] Individuals with blood group O are more likely to present with severe cholera than individuals with non-O blood groups.[46] Hypochlorhydria increases the likelihood and severity of disease.[47,48] Retinol deficiency is associated with an increased risk for cholera,[49,50] and concomitant intestinal parasitic and bacterial infection can modulate anti–*V. cholerae* immune responses after cholera.[51,52] First-degree relatives of patients with cholera also have greater odds of becoming infected than

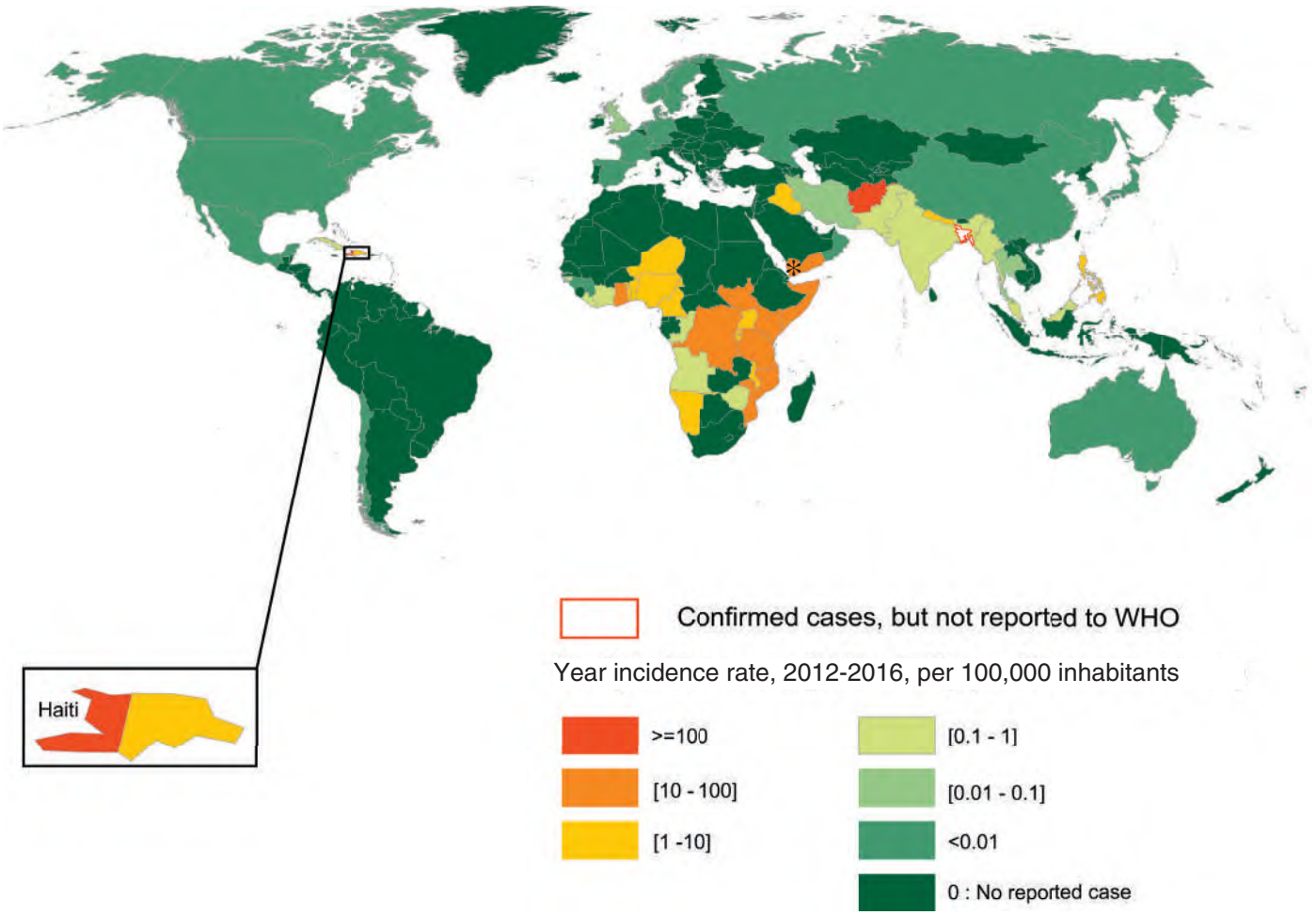

FIG. 214.1 Map of global distribution of annual incidence of cholera. Results represent mean annual incidence for the years 2014–2016. Data do not include large 2017 outbreak in Yemen *(asterisk). (Figure created by Martine Piarroux using Philcarto, http://philcarto.free.fr.)*

non–first-degree relatives in the same household, independent of blood group,[49] suggesting that other genetic factors may contribute to susceptibility, and in a candidate gene study a variant in the promoter region of *BPIFB1* (also known as *LPLUNC1*) involved in innate immune responses was associated with cholera.[53,54] In a genome-wide study, host susceptibility to cholera was also associated with potassium channel genes involved in cAMP-mediated Cl^- secretion and for components of the innate immune system involved in nuclear factor kappa B signaling.[55]

DIAGNOSIS

Cholera should be considered in any area of the world when an individual 5 years of age or older develops severe dehydration or dies as a result of acute watery diarrhea and when an individual 2 years of age or older develops acute watery diarrhea in an area of the world known to be endemic for cholera.[56] Microbiologic analyses of stool samples permit confirmation of *V. cholerae* and determination of antimicrobial susceptibility profiles; however, once cholera is known to be endemic in an area and the antimicrobial susceptibility profile of the outbreak strain is known, routine stool culture from each individual is not indicated. *V. cholerae* can be isolated from stool by using selective media such as taurocholate tellurite gelatin agar or thiosulfate citrate bile sucrose agar, and recovery of organisms can be enhanced by enrichment of stool in alkaline peptone water.[57] Serogroup and serotype identification are determined with specific antibodies.[58] Darkfield microscopy can be used to rapidly evaluate whether watery stools contain the characteristic "shooting star" motility of vibrios, which can be inhibited by the action of specific antibodies.[59] A number of immunoassays that can detect *V. cholerae* LPS or CT or both directly in stool have been developed.[60] These assays are particularly helpful when included in surveillance systems in areas of the world at risk for cholera. During laboratory evaluation and under field conditions, the sensitivity and specificity of these assays have ranged from 60% to 100%.[60] A commercially available PCR panel for detection of gastrointestinal pathogens in stool includes *Vibrio cholerae* (Biofire FilmArray).

TREATMENT

The primary focus of treating patients with cholera is fluid management. Patients with suspected cholera should be rapidly restored to euvolemia, and ongoing fluid losses need to be matched. Algorithms for rapid assessment of fluid status in any setting, including in sites lacking electricity or laboratory support, are shown in Table 214.1. Although patients with severe cholera or who are unresponsive require administration of intravenous fluids in the initial stages of treatment, many patients with less severe cholera and who are alert can be managed with oral rehydration therapy alone.

Management of patients with cholera does not usually require any laboratory analysis, although focused laboratory analysis can assist clinical decisions in complicated cases including in patients who do not respond to fluid replacement therapy or have ongoing coma, seizures, or severe ileus. In these cases, focused testing for persistent hypoglycemia, hyponatremia, hypernatremia, hypokalemia, hypocalcemia, renal dysfunction, and degree of hemoconcentration can assist in management.[61]

Patients with severe dehydration from cholera have greater than a 10% deficit of their total fluid volume and often require greater than 100 mL/kg of fluid via the intravenous route within 3 hours of presentation (6 hours in children younger than 1 year) to restore euvolemia, in addition to matching ongoing losses. Once patients are mentally alert, oral rehydration fluid should supplement administration of intravenous fluids. Intravenous therapy should be discontinued once the patient can maintain euvolemia through oral ingestion alone, usually within 3

TABLE 214.1 How to Approach Rehydration in Patients With Suspected Cholera

		DEGREE OF DEHYDRATION		
		None (<5%)	**Some (5%–10%)**	**Severe (>10%)**
Assess degree of dehydration	Mentation	Alert	Restless, irritable	Lethargic or unconscious
	Eyes	Normal	Sunken	Sunken
	Skin turgor	Normal recoil	Slow recoil	Very slow recoil (>2 s)
	Pulse	Normal	Rapid, low volume	Weak or absent
	Thirst	Drinks normally	Thirsty, drinks eagerly	Drinks poorly or unable to drink
Approach to rehydration[a]	Fluid replacement	Ongoing losses only	75 mL/kg in addition to ongoing losses	>100 mL/kg in addition to ongoing losses
	Preferred route of administration	Oral[b]	Oral or intravenous	Intravenous
	Timing	Usually guided by thirst	Replace fluids over 3–4 h	As rapidly as possible until circulation is restored; complete the remainder of fluids within 3 h
	Monitoring	Observe until assured ongoing losses can be adequately replaced by ORS	Observe every 1–2 h until all signs of dehydration resolve and patient urinates	Once circulation is established, monitor every 1–2 h

[a]Patients with comorbid conditions including severe malnutrition and significant complications, infants, and elderly patients may require adjustments from these standard guidelines for the inpatient treatment of severely malnourished children. See references 56, 63, and 77.
[b]If losses are greater than 10 mL/kg/h, it may not be possible to successfully initially use oral therapy.
The Cholera Outbreak Training and Shigellosis Program (www.cotsprogram.com) is an excellent resource that provides free online information regarding the management of patients with cholera based on World Health Organization standards.
ORS, Oral rehydration solution.
Modified from Harris JB, Larocque RC, Qadri F, et al. Cholera. Lancet. *2012;379:2466–2476.*

to 6 hours of presentation, and subsequent fluid support should be delivered solely through the oral route. If intravenous fluids are not available, an orogastric or nasogastric tube may be inserted in unresponsive patients to permit administration of appropriate oral fluid containing both glucose and sodium in equimolar amounts.

Patients with the most severe form of the disease, cholera gravis, may require on average 200 mL/kg of fluid replacement within the first 24 hours and may require as much as 350 mL/kg in this period.[56,61,62,63] Ongoing fluid losses may exceed 10 to 20 mL/kg/h, and output should be matched by fluid replacement. A cholera cot (Fig. 214.2) is a simple and inexpensive device that allows care workers to monitor fluid losses, enabling accurate matching fluid replacement. If a cholera cot is not available, fluid replacement should be guided by estimating 10 to 20 mL/kg of body weight for each diarrheal stool or episode of severe vomiting during cholera. Severely malnourished children with cholera require special clinical care.[64]

Patients with some dehydration often have a fluid deficit of 5% to 10% and may be irritable and restless but are not lethargic or unconscious. Euvolemia should be reestablished within 3 to 4 hours of presentation and often requires at least 75 mL/kg of fluid in addition to matching ongoing losses. Individuals with less than 5% dehydration will usually be mentally alert and able to drink normally and should largely be treated with oral fluid management.

When intravenously administered fluids are indicated, lactated Ringer solution (LRS), ideally supplemented with 5% dextrose (D5LR), is considered optimal among the most commonly available fluids.[61] D5LR contains not only sodium chloride and glucose but also potassium and bicarbonate to partially match ongoing losses, although the amount of potassium present in LRS is insufficient to match the ongoing losses associated with cholera (Table 214.2). Although normal saline can also be administered with the primary goal of maintaining circulatory support, the absence of potassium, bicarbonate, and glucose (unless supplemented) make it a less ideal agent. Normal saline with 5% dextrose should be used only if D5LR is not available, and D5LR should ideally be supplemented with additional potassium if available. In highly endemic areas, local manufacturers will sometimes produce cholera saline, often known as Dhaka solution, which contains increased potassium and bicarbonate and glucose compared with LRS and which was developed to optimally match fluid losses during cholera.[61]

Oral rehydration solutions (ORSs) are based on the delivery of equimolar concentrations of sodium and glucose in the small intestine to maximize absorption by relying on a different absorption pump than that affected by CT.[65] WHO initially recommended that patients with cholera be treated with an ORS with an electrolyte profile that closely matched that of cholera stools; however, with the broader use of ORS to prevent dehydration-associated deaths and morbidity from all causes of gastroenteritis, that formula has been modified to include less salt. This lower-osmolarity formula more closely reflects the electrolyte profile of stool associated with all-cause gastroenteritis globally. Lower osmolarity ORS limits the risk of hypernatremia in young children with non–cholera-associated dehydration, and although it increases the likelihood of subclinical hyponatremia in treated patients with cholera, this degree of hyponatremia does not appear to be clinically significant at a population level.[66] If available, rice-based ORS formulas can decrease stool losses in severe cholera and duration of diarrhea.[67] During emergency situations, ORS can be made by mixing ½ tsp of table salt with 6 tsp of table sugar in 1 L of safe water.[68] Potassium replacement can occur through ingesting locally available potassium sources such as coconut milk, bananas, or orange juice.

Unresponsive patients or patients with persistent hypoglycemia should receive 0.25 to 0.50 g/kg of intravenous glucose, and glucose should be continued until the patient is able to drink and eat to replenish glucose stores.[69] Potassium and bicarbonate levels should be evaluated in patients with severe or persistent ileus because these individuals may require supplemental potassium replacements. Rapid administration of intravenous fluids, especially fluids containing bicarbonate, can lead to calcium shifts that can precipitate tetany, which can be treated with intravenously administered calcium gluconate.

Despite the role that antibiotics play in many infectious diseases, their role in the treatment of a patient with cholera is secondary. Antibiotics should be administered to patients with moderate or severe dehydration from cholera.[70] The administration of effective antibiotics reduces the volume of diarrhea by approximately 50% and shortens the duration of diarrhea.[71,72] Because patients with moderate or severe dehydration from cholera consume the largest amounts of fluid in resource-limited settings, targeting antibiotic treatment to these individuals can maximize use of limited supplies. Appropriate antimicrobial therapy can also limit the shedding of organisms and may decrease the risk of ongoing transmission.[71,72] Increasing antimicrobial resistance in *V. cholerae* is occurring globally, and the seventh pandemic strains currently circulating contain an integrative conjugative element that mediates resistance to multiple antibiotics. Individuals with cholera who are treated with an antimicrobial agent should receive a macrolide such as azithromycin or a fluoroquinolone antibiotic based on local resistance profiles (Table 214.3). Resistance to tetracyclines is common, so these agents (including doxycycline) should be used only during outbreaks when antimicrobial susceptibility has been confirmed, and tetracyclines should, in general, not be used in pregnant women and children younger than 8 years.[73] Macrolide antibiotics are of particular use in the treatment of women of childbearing age and in young children.

Breastfeeding should be continued for infants with cholera in addition to other avenues of fluid replacement. To limit the impact of malnutrition

FIG. 214.2 (A) Buckets of rice water stool from a patient with cholera. (B) Cholera cots; note central hole and sleeve leading to buckets to assist with monitoring ongoing fluid losses. (C) Obtunded, severely dehydrated patient with cholera; note sunken eyes. (D) Same patient after rehydration therapy. *(From Chowdhury F, Khan AI, Faruque AS, et al. Severe, acute watery diarrhea in an adult.* PLoS Negl Trop Dis. *2010;4:e898; and Harris JB, Larocque RC, Qadri F, et al. Cholera.* Lancet. *2012;379:2466–2476.)*

TABLE 214.2 Composition of Cholera Stools and Therapeutic Fluids for Cholera

		CONCENTRATION (mmol/L)					
		Na^+	K^+	Cl^-	HCO_3^-	Carbohydrate	COMMENTS
Intravenous fluid	LRS	130	4	109	28	— (278 if D5LR available)	LRS contains potassium and bicarbonate and is preferred over NS. LRS and NS can be supplemented with dextrose. Dhaka solution more closely approximates losses during cholera, but is not readily available.
	NS	154	0	154	0	—	
	Cholera saline (Dhaka solution)	133	13	154	48	140	
Oral rehydration therapy	ORS (WHO, 2002)[a]	75	20	65	10 (citrate)	75 (glucose)	WHO ORS uses glucose as a carbohydrate source. Rice-based ORS formulations have been found in randomized trials to reduce duration of diarrhea and stool losses in severe cholera.[67] A homemade preparation of ORS could be used in an emergency situation.
	Rice-based ORS (e.g., CeraORS 75)	75	20	65	10 (citrate)	27 g rice syrup solids	
	Homemade ORS[68]: ½ tsp table salt and 6 tsp table sugar in 1 L of safe water	≈75	0	≈75	0	≈75	
Electrolyte losses in stools (composite estimates)	Cholera stool, adult	130	20	100	45	—	Mean maximal rate of purging in severe cholera may exceed 10 mL/kg/h. Sodium losses in cholera stools exceed those seen in other causes of diarrheal illness.
	Cholera stool, child	100	30	90	30	—	
	Noncholera stool, child (ETEC)	50	35	25	20	—	

[a]In 2002 WHO replaced its previous formulation of ORS with the current lower osmolarity formulation to reflect the broad use of ORS for treating dehydration from all-cause gastroenteritis.

Cl^-, Chloride ion; *D5LR,* 5% dextrose in lactated Ringer solution; *ETEC,* enterotoxigenic *Escherichia coli; HCO_3^-,* bicarbonate ion; *K^+,* potassium ion; *LRS,* lactated Ringer solution; *Na^+,* sodium ion; *NS,* normal saline; *ORS,* oral rehydration solution; *tsp,* teaspoon; *WHO,* World Health Organization.

See references 56, 77, and 143.

Modified from Harris JB, Larocque RC, Qadri F, et al. Cholera. Lancet. *2012;379:2466–2476.*

TABLE 214.3 Antimicrobial Options for Treating Patients With Cholera

CLASS	ANTIBIOTIC	PEDIATRIC DOSE[a]	ADULT DOSE	COMMENTS
Macrolides	Erythromycin Azithromycin	12.5 mg/kg/dose qid × 3 days 20 mg/kg × single dose	250 mg qid × 3 days 1 g × single dose	Single-dose azithromycin is often preferred therapy, especially in children, and has been shown to be more effective than ciprofloxacin in randomized trials in regions where reduced susceptibility to fluoroquinolones is common.[144,145] There are rare reports of macrolide resistance.
Fluoroquinolones	Ciprofloxacin	15 mg/kg/dose bid × 3 days	500 mg bid × 3 days	In highly susceptible strains, single-dose ciprofloxacin compares favorably against erythromycin[146] and doxycycline[147] in randomized trials. Reduced susceptibility to fluoroquinolones has become common in endemic areas and is associated with treatment failure.[144,148]
Tetracyclines	Tetracycline Doxycycline	12.5 mg/kg/dose qid × 3 days 4–6 mg/kg × single dose	500 mg qid × 3 days 300 mg × single dose	Antibiotic resistance to all tetracyclines is common.[73] Empirical use is often reserved for outbreaks caused by documented susceptible isolates. In general, tetracyclines are not recommended for pregnant women or children less than 8 years old.

[a]Pediatric doses are based on weight; should not exceed maximum adult dose.
bid, Twice a day; *qid,* four times a day.
Modified from Harris JB, Larocque RC, Qadri F, et al. Cholera. Lancet. *2012;379:2466–2476.*

and energy deprivation, a high-energy diet should be encouraged as soon as fluid status is recovered for all individuals with cholera. WHO recommends zinc for children with diarrhea who are younger than 5 years, regardless of cause; zinc supplementation has been associated with a decreased volume and duration of diarrhea in children with cholera.[74] Zinc supplementation is usually given as 20 mg/day for 10 days for children 6 months of age to 5 years and 10 mg/day for 10 days for children younger than 6 months.[56] Vomiting of the supplement by young children is common during the initial stages of fluid resuscitation, but ingestion should be encouraged. Although not specifically studied with cholera, vitamin A supplementation is recommended for children with diarrhea in resource-limited settings. Although development of an antisecretory drug could theoretically limit fluid losses during cholera, no such agent has yet been identified and developed for widespread use. Antimotility agents should be avoided during cholera because they could lead to intestinal pooling of fluid (but not decreased production of fluid), yielding a false sense of improvement, and they can impede the ability to monitor ongoing fluid losses. Individuals caring for patients with cholera, who often include family members in resource-limited settings, should be carefully instructed that diarrhea will persist and may even worsen once an affected individual becomes rehydrated. Diarrhea usually resolves over 1 to 3 days.

Most patients who die of cholera die as a result of severe dehydration. The most common clinical mistakes in caring for patients with cholera are (1) inadequate initial fluid resuscitation; (2) inadequate matching of ongoing fluid losses, especially in young children; and (3) use of suboptimal or incorrect fluids.[75] With proper fluid management, mortality from cholera in patients who are able to reach clinical care should be less than 1%. However, many individuals die of cholera before they reach medical care, often within 6 to 12 hours of onset of symptoms, and case-fatality rates of 2% to 10% in the initial stages of complex emergencies are not infrequent.[76] Establishment of cholera treatment centers within communities with cholera outbreaks facilitates rapid access to care. Resources are available to assist in handling cholera outbreaks including the Cholera Outbreak Training and Shigellosis Program, an online resource that provides information, tools, and algorithms for the treatment of individuals and populations with cholera (www.cotsprogram.com).[77]

The treatment of a population with cholera, as opposed to an individual patient, can significantly strain limited resources, staff, and infrastructure. Cholera epidemics can cause panic in a population and in care providers who are unfamiliar with the disease. At-risk populations should be informed through community announcements, text messaging, the Internet, radio announcements, placards, posters, and other outreach programs as well as by local community leaders and personalities to recognize the initial symptoms of cholera, the importance of fluid replacement, and the need to seek medical care early in the illness. Simultaneous outreach programs should focus on the importance of safe water and adequate sanitation, and delivery efforts should focus on the provision of safe water and adequate sanitation including distribution of chlorine tablets for point-of-use treatment of unsafe water, if indicated. Integration of cholera vaccination programs should also be considered.

A community response to a cholera outbreak requires closely coordinated efforts that should involve community leaders as well as local, regional, national, and international health authorities. The unacceptably high case-fatality rates that are common in the initial phases of cholera outbreaks underscore the difficulty of establishing such coordinated efforts in the displaced, disenfranchised, and impoverished populations that are frequently most at risk for cholera as well as the often very complex nature of the emergencies that can afflict such populations.

IMMUNE RESPONSES

Although the genomes of *V. cholerae* O139 and El Tor O1 are extremely similar, including containing identical CT genes,[31,78] infection with *V. cholerae* O1 provides little to no protection against *V. cholerae* O139 and vice versa.[58] Similarly, although immune responses targeting CT are common after cholera, they do not mediate long-lived protective immunity.[79] As such, protective immunity against cholera appears to be largely serogroup specific and toxin independent, and serogroup specificity is determined by the OSP of LPS.[80]

Historically the vibriocidal response has been commonly used as a gauge of protective immunity against cholera.[81] This assay measures a complement-dependent bactericidal antibody response in blood that presumably is a surrogate marker for as yet poorly understood mucosal responses.[82] Every twofold increase in the vibriocidal response is associated with an approximately 40% reduction in the risk of cholera in Bangladesh.[83] The vibriocidal response is largely composed of immunoglobulin M antibodies targeting LPS, and anti-LPS responses (and anti-LPS memory B cells) have been associated with protection against cholera in household contacts of index cholera patients in Bangladesh.[49,84] OSP and LPS responses highly correlate, and vibriocidal responses are adsorbed away by OSP, suggesting the importance of OSP in mediating protective immunity to cholera.[80,85] Indeed, OSP-specific plasma and memory B-cell responses are associated with protection against cholera in household contacts of cholera index patients, and OSP-specific antibodies are associated with protection against cholera in North American volunteers experimentally infected with virulent *V. cholerae*.[85a,85b] T-cell activation during cholera is also associated with induction of memory B-cell responses,[86] and activation of innate immune responses during acute cholera correlates with subsequent immune responses.[87,88]

Analysis of the humoral response against *V. cholerae* suggests that the vast majority of the responses target a relatively small number of immunodominant antigens including OSP, CT, and sialidase, a bacterial enzyme that potentiates toxin activity.[89,90] Young children develop immune responses after cholera that are comparable to responses that develop in older children and adults including to antigens considered T-cell independent such as LPS and OSP.[91,92] The mechanism by which young

children surviving cholera develop such responses is unclear, but the potent adjuvant activity of CT may contribute.[91,92]

PREVENTION AND VACCINES

Prevention or eradication of cholera from the world is extremely laudable and a potentially achievable goal. However, because *V. cholerae* has an environmental reservoir, complete eradication of cholera from the world will be a very difficult task. Control of epidemic cholera has already been achieved in many countries. In the United States and Northern Europe, epidemic cholera was eliminated before the wide use of antibiotics and vaccines through the provision of safe water and adequate sanitation. When such basic public health measures are available globally, epidemic cholera will likely be eliminated.[93] This is a daunting goal; WHO currently estimates that more than 800 million individuals currently lack safe water and that more than 2 billion individuals currently lack adequate sanitation.[94] In light of these realities and the projected population growth over the next few decades, especially in densely populated regions with limited infrastructure, it is likely that human populations will be at risk for cholera for decades.[95] Indeed, the number of countries endemic for cholera and the number of outbreaks, their duration, their rate of recurrence, and their associated case-fatality rates all have increased during the current pandemic that began in 1961.[76,95]

Because the global provision of safe water and adequate sanitation will be extremely costly and take many years to achieve, integrated control programs should in many situations include use of cholera vaccines. The development of both oral and parenteral cholera vaccines has a long history, dating back to the late 1800s. A killed whole-cell parenteral cholera vaccine was often required for international travel for much of the mid-1900s. This vaccine had only modest short-term protective efficacy, required two to three immunizations as a primary series, required booster immunizations every 6 months, and had a high frequency of adverse events.[96] The vaccine eventually fell out of favor, in part because of the perceived decreased risk for routine international travelers of acquiring cholera and the impracticality of its ongoing use among populations in areas endemic for cholera. The shortcomings of this vaccine were part of the calculus that led to the removal of cholera vaccines from public health responses to control cholera in the latter part of the 1900s.

This calculus changed in light of the prolonged and worsening ongoing pandemic and the development of safe, better tolerated, and more effective cholera vaccines that induce longer-term immunity.[82] A number of oral killed cholera vaccines approved by WHO are currently commercially available internationally (Table 214.4).[39] One, Dukoral (Valneva, Lyon, France), a whole-cell/recombinant–B subunit oral cholera vaccine, contains approximately 10^{13} killed *V. cholerae* O1 classic and El Tor organisms and is supplemented by 1 mg of the nontoxic B subunit of cholera toxin (CtxB). Another vaccine, Shanchol (Shantha Biotechnics–Sanofi Pasteur, Hyderabad, India), is bivalent and includes a number of *V. cholerae* O1 strains as well as *V. cholerae* O139; it is not supplemented by CtxB. A related vaccine is the oral cholera prevention vaccine Euvichol (Eubiologics, Republic of Korea); Euvichol is now the primary vaccine in the WHO cholera vaccine global stockpile.[97] A locally produced Vietnamese version of the bivalent vaccine that is not internationally available is mORCVAX (VaBiotech, Hanoi, Vietnam).

The three prequalified oral cholera vaccines (Euvichol, Shanchol, Dukoral) are safe and well tolerated.[98–112] Dukoral is approved for use in children 2 years of age and older; Shanchol and Euvichol are approved for use in children 1 year of age and older. The absence of supplemental CtxB from Shanchol and Euvichol removes the requirement for buffer and decreases the amount of fluid that must be administered at the time of immunization. The vaccines are administered as two or three oral immunizations and provide approximately 60% to 80% protective efficacy for 6 to 60 months, depending on the vaccine used and age of the recipient. Booster immunizations are currently recommended with Dukoral every 6 months for children younger than 5 years of age and after 2 years for all other recipients. Shanchol may be associated with longer-term protection, although its protective efficacy in children younger than 5 years is also of lower magnitude and shorter duration than that afforded to older recipients.[113]

The manufacturers of Shanchol and Euvichol do not currently have recommendations regarding revaccination.[39] At the present time, the manufacturers do not recommend administration of either vaccine to pregnant women, although the scientific basis for this recommendation is unclear because the vaccines contain only killed organisms; the vaccines are not systemically administered or absorbed; and pregnant women who develop cholera are at significant risk of severe dehydration, clinical complications, and loss of the pregnancy. Administration of Dukoral was not associated with an increased risk of adverse birth defects or sequelae among pregnant women who inadvertently received the vaccine.[114] Shanchol has also been safely administered in populations with a high prevalence rate of human immunodeficiency virus.[111,115] Oral killed cholera vaccines may not require a cold chain and have shown some, albeit reduced, efficacy after a single dose.[116,117,118]

Modeling suggests that vaccine coverage rates of 50% or higher can potentially interrupt transmission secondary to vaccine herd affect[119,120] and that the earlier a cholera vaccine rollout program occurs during an outbreak, the more significant the impact of immunization.[121–123,124,125] Some studies have found cholera vaccine use to be cost-effective, whereas others have not.[126,127]

In light of the safety, immunogenicity, protective efficacy, and potential impact of cholera vaccines, the WHO Cholera Control Task Force now recommends that cholera vaccines be considered as part of an integrated cholera control program in areas endemic for cholera, in humanitarian crises with high risk of cholera, and during cholera outbreaks.[39] In

TABLE 214.4 Internationally Available and WHO Prequalified Oral Killed Cholera Vaccines

VACCINE	DOSES[a]	DOSING INTERVAL[a]	DOSING VOLUME[a]	BOOSTERS[a]	PROTECTIVE EFFICACY	COMMENTS
Shanchol (Shantha Biotechnics–Sanofi Pasteur); Euvichol (Eubiologics)						
≥1 year of age	2	14 days (7–42 days probably permissible)	1.5 mL	No recommendations from manufacturer	60%–70% protective efficacy decreasing to baseline over 36–60 months in older children and adults; 40% protective efficacy and shorter duration in younger children	Appears to provide longer-term protection than that provided by Dukoral. Does not require buffer to administer vaccine. Maintained in WHO Global Cholera Vaccine Stockpile
Dukoral[b] (Valneva)						
Children 2 to <6 years of age	3	14 days (7–42 days permissible)	3 mL vaccine and 75 mL buffer	Every 6 months	60%–85% protective efficacy within 6 months of vaccination, decreasing to baseline over 24–36 months	Licensed in many countries. Provides short-term protection against diarrhea caused by heat-labile toxin expressing strains of ETEC.
≥6 years of age	2	14 days (7–42 days permissible)	3 mL vaccine and 150 mL buffer	Every 2 years		

[a]Per manufacturer.
[b]Field studies have involved both the current preparation of whole-cell vaccine supplemented with recombinant cholera toxin B subunit and an initial preparation of whole-cell vaccine containing nonrecombinant B subunit.
ETEC, Enterotoxigenic *Escherichia coli; WHO,* World Health Organization.