pathognomonic for CDI and may also occur in antibiotic-associated diarrhea not caused by *C. difficile.*[24,25] Symptoms may occur while patients are receiving antibiotics, usually after 5 to 10 days of therapy, or can occur 2 to 10 weeks after antibiotic therapy has been completed.[26] All classes of antibiotics have been associated with CDI, including the penicillins, cephalosporins, macrolides, lincosamides, and aminoglycosides. Diarrhea accompanied by fever occurs in most patients and resembles the symptoms caused by many other intestinal pathogens. In severe cases, bloody diarrhea may be present. Diarrhea is often accompanied by bloating and cramping. Leukocytosis is not uncommon. CDI is more common in the elderly and in hospitalized patients receiving broad-spectrum antibiotic therapy. When surgical intervention is required for severe disease, colostomy carries a substantial risk of mortality regardless of the age of the patient.[27]

Molecular Pathogenesis

The initiating event for CDI involves the disruption of the intestinal microbiome and the normal mechanisms controlling clostridial populations for clostridia such as deconjugated bile acids during treatment with antibiotics. While it is not uncommon for mild diarrhea to occur simply due to the disruption of the microbiome when broad-spectrum antibiotics are used, the presence of *C. difficile* in the intestine can lead to more serious toxin-mediated disease. Since *C. difficile* spores can survive even high concentrations of antibiotics, germination of spores and rapid growth of the organism may occur as antibiotic levels in the lumen of the bowel decrease below inhibitory concentrations for *C. difficile* prior to the recovery of the normal microbiome.

The virulence of *C. difficile* is due to production of two high-molecular-weight toxins, *C. difficile* toxin A (TcdA) and *C. difficile* toxin B (TcdB), originally described as an enterotoxin and a cytotoxin, respectively. Intestinal epithelial cells endocytose the toxins and, once internalized, the toxins irreversibly glucosylate members of the Rho family of small GTP-binding proteins. As a result, there are major alterations in the actin cytoskeleton. These changes lead to the characteristic cytotoxic effects noted in cell culture by compromising the cell's ability to internalize nutrients and carry out vesicular transport, both of which may lead to cell death. The genes for TcdA, TcdB, and an RNA polymerase sigma factor, *C. difficile* toxin R (TcdR), are carried as a pathogenicity locus. In addition, *tcdC,* a gene that downregulates toxin production by interfering with TcdR, is part of this locus.[5,28–30] Hypervirulent toxin-producing strains, with a deletion within the *C. difficile* toxin C gene (*tcdC*), have been reported and account for the current concerns about more virulent CDI. Deletions in the *tcdC* gene can contribute to unregulated toxin production, resulting in much higher levels of toxin being produced. A particular outbreak-associated ribotype, O27/NAP1, is attributed with causing many cases of severe disease. Alterations in the *tcdC* gene have been noted in multiple ribotypes, with the O27 ribotype currently the most commonly associated with *tcdC* deletions.[31] In addition to the two major toxins TcdA and TcdB, a binary ADP ribosylating toxin—*C. difficile* toxin (CDT), a common A-B toxin motif for many clostridial species—has been reported to contribute to the occurrence of more severe CDI.[31–34] Not all strains of *C. difficile* produce all three toxins (TcdA, TcdB, and CDT). Surveillance studies indicate that production of both TcdA and TcdB is most common, occurring in about 65% of strains, with TcdB (cytotoxin) produced by 97% of all strains evaluated. This likely accounts for the utility of the cytotoxicity assay as a diagnostic method before the full range of toxins produced by *C. difficile* was appreciated.

Treatment and Diagnosis

Less serious presentations of CDI have been treated with metronidazole since the late 1990s, with vancomycin reserved for treatment of serious and/or progressive disease.[35] However, given the occurrence of rapidly progressing severe disease in some patients, vancomycin or fidaxomicin (see later in this chapter and see Chapter 243) may be used in cases of moderate disease.[36] Adjunct therapies using probiotics to reconstitute the normal microbiome of the intestine or anion exchange resins that bind toxin have been proposed, but appear to be of limited value and may be harmful for the patient.[35,37]

Of particular concern is the recurrence of CDI once appropriate therapy has been discontinued. This is thought to be due to the survival of the biologically inert spores during therapy and regrowth of the vegetative cells once therapy is discontinued in a microbiologic environment that has been damaged by CDI and by the subsequent antibiotic therapy. It has been shown in gnotobiotic mice that the concentrations of *C. difficile* can be suppressed by vancomycin, but that low levels of spores survived during treatment. The concentration of *C. difficile* returned to pretreatment levels once therapy was discontinued, indicating that the spores survived antibiotic treatment and germinated when antibiotic levels declined.[38] A relatively new antibiotic—fidaxomicin, an RNA polymerase inhibitor—has been shown in clinical trials to reduce the rate of recurrent CDI by almost 50%.[39–41]

Approximately 20% of patients treated for CDI develop recurrent disease. Current recommendations include repeating initial therapy. For a second recurrence, a pulsed dosing of vancomycin is recommended; however, the successful use of fecal microbiota transplants is increasingly becoming the treatment of choice for multiple recurrences of CDI.[36,42–46] Published reports suggest a greater than 90% efficacy in patients with recurrent disease.[47] There are currently a number of ongoing clinical trials of fecal microbiota transplantation for various inflammatory bowel diseases.[47–51] Alternatives to fecal microbiota transplantation and fidaxomicin for recurrent disease also include the use of bezlotoxumab, a monoclonal antibody directed at TcdB during primary CDI to prevent recurrent disease.[52]

Laboratory diagnosis of CDI is generally accomplished by detection of toxin in the stool specimens of patients suspected of having clinical disease. While culture can be performed to isolate *C. difficile,* the presence of the organism does not necessarily mean CDI is present because 4% to 10% of overtly healthy adults may carry this organism as part of their normal intestinal microbiome, and simply detecting the organism by antigen, toxin gene polymerase chain reaction (PCR), or "toxigenic culture" does not necessarily correlate with clinical disease. Initially, the cytotoxicity assay for TcdB was used to diagnose CDI. In this assay, a monolayer cell culture is exposed to a filtrate of stool for 24 to 48 hours and examined for the characteristic cytopathic effect of TcdB. If detected, a portion of the filtrate is mixed with antibody to TcdB and a second cell culture used to determine if the cytopathic effect can be neutralized (Fig. 246.2). While this is a very sensitive assay, the time required to make a definitive diagnosis is often 48 to 72 hours. Currently, there are a variety of enzyme immunoassays (EIAs) available to provide rapid detection of TcdA, TcdB, or both. Assay sensitivity ranges from less than 50% to better than 93%, depending on the assay used and the quality of the specimen, while assay specificity is usually greater than 90%.[53,54] Most EIAs include detection of both TcdA and TcdB, since it is known that some strains produce one toxin but not the other. While the prevalence of CDI varies depending on patient demographics, one large Boston teaching hospital performed over 8000 EIAs in 2012, with over 10% yielding positive results for one or both toxins (unpublished data).

PCR assays designed to detect the genes have also become popular for detection of CDI. However, these assays detect one or more of the toxin genes, but detection of the toxin genes does not necessarily mean that toxin is being produced. Toxin production can only be measured by looking directly for the toxins or the transcription products of the toxin genes *tcdA* and *tcdB.* The current algorithm used in many diagnostic laboratories includes preliminary testing for an antigen, glutamate dehydrogenase (GDH), produced by most strains of *C. difficile* and the presence of toxin using a membrane immunoassay. If antigen is present but no toxin is detected, a PCR assay that detects *tcdA* and *tcdB* is currently recommended to confirm CDI. If the PCR assay is positive, then it is presumed that a symptomatic patient has CDI. This approach is likely to change because the PCR assay only confirms that *C. difficile* is present, but not that toxin is being produced. Many patients positive for GDH and PCR with negative results for toxin are being treated for CDI when the cause of their disease may be another agent/toxin that is not being assayed. Recent studies in England have pointed out the problems with the existing algorithm employed in the United States and note that the best clinical correlation in symptomatic patients is to test for *C. difficile* with either GDH or PCR testing and a sensitive toxin assay.[55,56]

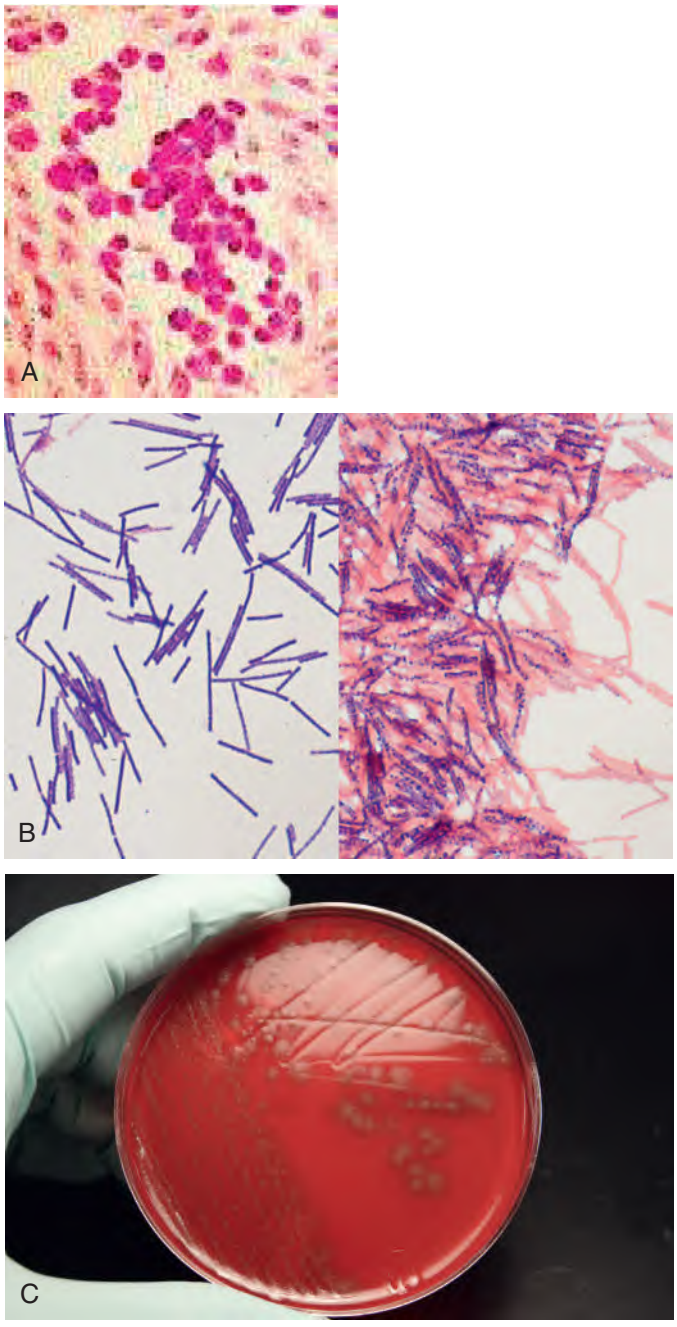

FIG. 246.2 *Clostridioides difficile* (formerly *Clostridium difficile*). (A) Cytopathic effect caused by *C. difficile* toxin B (TcdB). (B) Gram stain of *C. difficile*. (C) Growth of *C. difficile* on blood agar.

While *C. difficile* is responsible for many cases of antibiotic-associated diarrhea, it is not the only cause. Other clostridial species, such as *C. perfringens* and *Clostridium sordellii,* are also capable of producing enterotoxins under similar circumstances that lead to diarrheal disease during antibiotic therapy.[25] Therefore a negative assay for *C. difficile* toxins does not preclude the involvement of other clostridial species in the clinical manifestations.

When cultured on selective agar media, *C. difficile* can be readily isolated from stool samples (see Fig. 246.2). The colonies are generally flat, 4 to 6 mm in diameter, and gray in color, with irregular margins and a ground-glass appearance. A distinctive "barnyard" odor is often detected from cultures grown on blood agar media because of production of *p*-cresol. Gram stain of colony growth usually shows gram-positive to gram-variable, somewhat slender rods, with subterminal spores when present. Definitive identification requires both fermentation profiles and analysis of short-chain fatty acid end products of fermentation or the use of 16S rDNA sequencing.

It is extremely important for appropriate infection control measures to be implemented within the hospital or nursing home environment for patients diagnosed with CDI. These measures include the rapid diagnosis of CDI in patients on antibiotic therapy, contact precautions for patients with CDI, appropriate therapy, and rigorous cleaning of rooms between patient admissions using disinfectants that effectively kill spores.[6]

CDI: an Emerging Public Health Crisis

The incidence of hospital-based CDI diagnoses has skyrocketed. There was a 117% increase in the listing of CDI on hospital discharges in the Healthcare Cost and Utilization Project website from 2000 to 2005.[57,58] CDI-related death rates have steadily increased as well over the past decade. Redelings and colleagues reported a 35% increase in mortality rates from 1999 (5.7 per million US population) to 2004 (23.7 per million US population).[59] Similar mortality trends have been observed in Great Britain.[60] Numerous recent studies underscore the importance of the observation that CDI represents a major public health concern requiring increased prevention, surveillance, and reporting.[61–64]

Clostridium perfringens and Clostridial Myonecrosis (Gas Gangrene)

Clostridial myonecrosis, or gas gangrene, is most often caused by a traumatic injury that becomes contaminated with clostridial spores from species that cause myonecrosis, most commonly those of *C. perfringens.* Throughout history, cases of gas gangrene have increased during times of human conflict due to the numbers of traumatic injuries provoked during violent conflict. Although other clostridial species can and do cause gas gangrene, the prevalence of *C. perfringens* in such infections marks it as one of the major clostridial pathogens. *Clostridium perfringens* can be isolated from soil and sediment samples from any geographic region. It is also one of the most common clostridial species isolated from the intestinal tract of humans and other animals. The ability of *C. perfringens* to cause myonecrosis is due to the production of a variety of protein toxins. Since strains of *C. perfringens* vary in their ability to produce some of the major toxins, this attribute differentiates this species into strain types (Table 246.2). Epidemiologically, *C. perfringens* type A is most common in human stool,[65] with other types more commonly associated with food poisoning and other enterotoxigenic disease (see later). Gas gangrene, while most often associated with *C. perfringens,* can be caused by other *Clostridium* species, including *C. septicum, C. sordellii, C. novyi, C. bifermentans,* and *C. histolyticum.* The common denominator for these species is the production of exotoxins, most often lecithinase, that devitalize tissue and promote invasive disease and myonecrosis.

TABLE 246.2 Toxins Produced by *C. perfringens*

TOXIN	STRAIN TYPES	BIOLOGIC ACTIVITY
α	All strains	Lecithinase
β	B and C	Necrotoxin, necrosis of the bowel
ε	B and D	Lethal, hemorrhagic
ι	E	ADP ribosylating; lethal
cpe	A, C, and D	Cytopathic
Neuraminidase	All strains	Hydrolyses *N*-acetylneuraminic acid
δ	B and C	Hemolysins
κ	All strains	Collagenase
λ	B, D, and E	Protease
μ	All strains	Hyaluronidase
ν	All strains	DNase

cpe, C. perfringens enterotoxin.

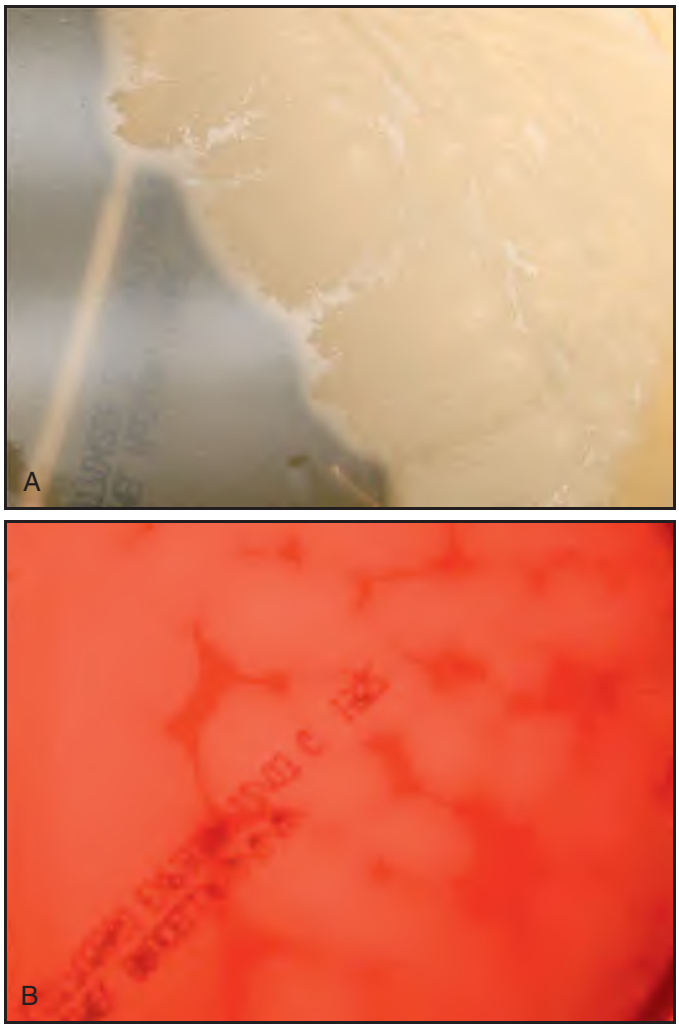

FIG. 246.3 ***Clostridium perfringens.*** (A) Lecithinase production by *C. perfringens* on egg yolk agar. Note the opaque zone surrounding the colony. (B) Hemolysin production by *C. perfringens* on blood agar.

Pathogenesis

All strains of *C. perfringens* produce α-toxin, a lecithinase (Fig. 246.3) that causes damage to cell membranes. During severe sepsis with *C. perfringens,* rapid hemolytic anemia may occur due to hemolysis caused by the α-toxin. The α-toxin is considered the major lethal toxin produced by *C. perfringens* during gas gangrene. In addition to the α-toxin, most strains produce additional hemolysins, proteases, collagenase, hyaluronidase, DNase, and neuraminidase.

Gas gangrene is most common following traumatic injuries that result in lowered tissue oxygen tension, such as crushing or penetrating injuries; the presence of foreign bodies, including soil or pieces of the object causing penetrating trauma; and mixed infections containing other organisms capable of reducing the oxygen levels at the site of infection. Studies of wounds occurring on the battlefield indicate that most are contaminated by clostridial spores, yet only a small percentage of these contaminated wounds result in clostridial myonecrosis. In the civilian population, approximately 10% of crushing wounds occurring as a result of automobile accidents have been shown to contain clostridial spores.[66] Clostridial myonecrosis may also occur following surgery, most often of the gastrointestinal (GI) or biliary tract, and following septic abortions. Contamination of lesions in which vascular insufficiency is present (e.g., diabetic foot ulcers), the occurrence of damaged tissue associated with burns, and underlying neoplastic disease may also contribute to the occurrence of gas gangrene.[67] Infections of the central nervous system are rare, but do occur.[68]

Diagnosis and Treatment

Clinically, clostridial myonecrosis generally begins within 24 to 72 hours after traumatic injury or surgery. Initial symptoms may include severe pain in the absence of obvious physical findings, suggesting a deep tissue infection. When traumatic injuries penetrate the skin, redness at the site of the wound, followed by a rapidly spreading brown to purple discoloration of the skin, is often seen. The progression of gas gangrene is rapid, and within hours of the initial symptoms, edema and gas may be detected within the underlying tissues by physical examination, ultrasound, or radiographic evaluation. Hemorrhagic bullae may occur, along with a serosanguineous discharge and characteristic odor often described as "mousy." The odor is distinct from the putrid odor, due to the production of volatile amines, associated with gram-negative anaerobic infections. Gram staining of the discharge often reveals the typical gram-positive boxcar-shaped rods characteristic of *C. perfringens* (see Fig. 246.1A). Neutrophils are often not seen on Gram stain due to the lethal nature of α-toxin for these polymorphonuclear cells.[69] Gas gangrene leads to profound ischemia resulting from obstructed microvascular circulation caused by platelet aggregation within vessels and fibrin deposition. Unlike most soft tissue infections, in which the inflammatory process increases blood flow, lesions resulting from clostridial myonecrosis do not bleed readily.[70] These proischemic properties are attributed to the α-toxins of *C. perfringens* and *C. septicum,* both of which have similar biologic activity. The θ-toxin of *C. perfringens,* perfringolysin O, may also act synergistically with α-toxin to decrease microvascular perfusion. The toxins can also result in systemic hematologic derangements such as disseminated intravascular coagulation. Fever is often minimal during the early stages of disease; however, progression to full-blown sepsis with its classic hypotension, renal failure, and metabolic acidosis may set in rapidly.

Rapid diagnosis of clostridial myonecrosis, a true medical emergency, is critical for proper therapeutic intervention. Clinical signs such as severe pain at the site of the traumatic injury, tachycardia in the absence of fever and obvious systemic toxicity accompanied by edema, discoloration of the skin, appearance of hemorrhagic bullae, and gas detected within the tissues are typical findings. Gram staining of fluids or exudates showing the typical boxcar-shaped gram-positive rods and few polymorphonuclear cells is often the earliest laboratory findings. Spores are not seen in Gram stains of clinical specimens. Growth of *C. perfringens* on blood agar plates is rapid, with colonies often detected within 12 to 16 hours of inoculation. Colonies are yellowish to gray and opaque, and 4 to 8 mm in diameter with irregular borders, and exhibit a characteristic double zone of hemolysis with an inner zone of β-hemolysis surrounded by an outer zone of partial hemolysis (see Fig. 246.3B). The lecithinase or α-toxin can be detected by growth on egg yolk agar medium, which is rich in triglycerides (see Fig. 246.3A). An obvious white precipitation surrounding colonies is evidence of lecithinase production. Neutralization of this reaction by specific antisera (the Nagler reaction) is presumptive evidence for the identification of *C. perfringens.* Most clinical microbiology laboratories use a combination of fermentation reactions and detection of short-chain fatty acid end products for definitive identification of *Clostridium* spp. Although not widely used at present by clinical microbiology laboratories, rapid PCR methods for identification of *C. perfringens* are available.[71] Methods such as MALDI-TOF mass spectrometry may be used for rapid identification of organisms, including *Clostridium* spp.[12,72–75]

The most important part of therapy for clostridial myonecrosis is emergent surgical débridement of infected tissues. When the abdominal wall is involved, débridement of the affected muscle with generous resection margins of apparently healthy tissue is important to prevent recurrence or progression of infection. For extremities, amputation or extensive débridement is appropriate. Uterine gas gangrene most often requires emergency hysterectomy. Early antibiotic intervention is essential for survival, and penicillin remains the most commonly employed antibiotic.[76] Although there is some concern regarding the continued susceptibility of *C. perfringens* to penicillin, most strains tested are susceptible to easily achieved levels. Other agents, including metronidazole, clindamycin, and the carbapenems, can also be used for treating clostridial myonecrosis. *Clostridium perfringens* remains susceptible to most front-line antimicrobial agents, although resistance to antibiotics

such as clindamycin has been reported.[77] The role of hyperbaric oxygen treatment as adjunctive therapy remains controversial,[76,78] as a result of the nature of the compromised tissue perfusion attributable to clostridial-driven microvascular damage. Some clinicians argue that hyperbaric oxygen treatment makes it easier to identify viable tissue, which decreases the need for more extensive surgical débridement. Other adjunctive therapy includes the use of granulocyte colony-stimulating factor to stimulate hematopoietic proliferation. While some cases of clostridial myonecrosis have been successfully treated with surgical débridement and hyperbaric oxygen alone, inclusion of aggressive antibiotic therapy early in the course of treatment increases survival rates, particularly when combined with aggressive and early surgical débridement, and is considered the best therapeutic intervention.[76]

Clostridium botulinum and Botulism

Botulism refers to a group of organisms that all can produce one or more potent neurotoxins (BoNT) that cause paralysis. *Clostridium botulinum* is the most common species associated with botulism, but strains of other *Clostridium* species, including *C. baratii, C. butyricum,* and *C. argentinense,* have been shown to produce BoNT as well.

Botulism occurs as one of four clinical syndromes: foodborne intoxication, infant botulism, wound infection/intoxication, and intestinal colonization/intoxication in adults. According to the Centers for Disease Control and Prevention, there were 199 reported cases of botulism in the United States in 2015, the latest year for which data are available. Of these cases, 141 were infant botulism, 15 were wound infections, and 39 were food related. The major differentiating characteristic among strains producing BoNT is the serologic group of the neurotoxin produced (Table 246.3). Botulism in humans is caused by serotypes A, B, and E almost exclusively. Toxin serotypes C and D have been associated with disease in domestic and wild animals. Rarely, disease caused by types C, D, and F have been reported in humans. Strains that produce toxin type G are phenotypically different from other *C. botulinum* strains. Strains producing serotype G toxin have therefore been identified as *C. argentinense.* No cases of human disease due to serotype G toxin have been reported to date.

Infant botulism was first recognized in the United States in 1976, but has now been reported throughout the world in countries that have reporting systems for botulism.[79,80] Originally associated with the addition of honey to baby formula, there are now other recognized sources of spores associated with infant botulism.[81,82] The serotype distribution for these cases is similar to the serotypes that can be isolated from the soil of specific geographic areas. In the United States, approximately half of reported cases of infant botulism are from California, one of the states with an aggressive program for recognition and treatment of infant botulism. The disease results from the germination of ingested spores within the GI tract, with vegetative cells rapidly proliferating and producing BoNT, which is then absorbed and distributed via the peripheral circulation. Most cases occur in infants between 3 weeks and 6 months of age; however, cases have been reported to occur in infants a few hours after birth. Symptoms are like those described for adults, but may be harder to discern initially. Often constipation followed by lethargy and difficulty in feeding are the earliest signs of distress. There appears to be a spectrum of disease severity ranging from self-resolving cases that do not require treatment to life-threatening intoxications requiring hospitalization, supportive care, and specific antitoxin therapy. Treatment of infant botulism with human immune globulin that neutralizes BoNT (BabyBIG) has been approved by the FDA.[10]

Wound botulism occurs almost exclusively in users of illicit injectable drugs. Many of the reported cases involve the subcutaneous injection of drugs ("skin popping"), with a subsequent local infection with BoNT-producing clostridia. Absorption of BoNT into the peripheral circulation results in the flaccid paralysis characteristic of botulism.

Foodborne botulism occurs when BoNT is ingested with contaminated food. Historically, foodborne botulism was associated with improperly preserved meat products and canned foods. Today, foodborne botulism is extremely rare in the United States. Most current cases still occur as a result of ingestion of improperly prepared home-canned foods or sausage.

Adult intestinal botulism is similar to infant botulism in that ingested spores of BoNT producing clostridial species germinate and the vegetative cells proliferate and produce toxin that is absorbed by the host, resulting in characteristic symptoms. This particular form of botulism is exceedingly rare, with only an occasional report of affected adults.

Botulism is a paralytic disease process affecting the nerves stimulating skeletal muscle contraction. All BoNT serotypes block the release of acetylcholine at the neuromuscular junction, resulting in blockage of innervation of muscle activity. All serotypes of BoNT are heterodimeric proteins of approximately 150 kDa in size. In the native state, BoNT often is associated with other nontoxic proteins that serve to stabilize the toxin molecule. Once absorbed from the GI tract, the neurotoxin is rapidly distributed via the peripheral circulation. BoNT crosses the plasma membrane by receptor-mediated endocytosis. The light chain (molecular weight, 50 kDa), the catalytic domain of the molecule, is internalized into the nerve cell through a protein channel. BoNT serotype A is a zinc-dependent metalloprotease that blocks the release of acetylcholine by cleaving synaptosomal-associated protein of 25 kDa (SNAP-25), a soluble N-ethylmaleimide-sensitive factor attachment protein receptor (SNARE) protein necessary for exocytosis of this neurotransmitter.[10,83]

Botulism classically presents as an acute flaccid paralysis beginning with impairment of the cranial nerves that innervate the eyes, head, face, and pharynx. Paralysis descends in a symmetric fashion to involve the muscles of the thorax and extremities. When ingested, BoNT is often associated with GI symptoms, including abdominal cramps, nausea, vomiting, and diarrhea. In some cases, constipation may precede the descending paralysis. Death may result from paralysis of the tongue and pharyngeal muscles, leading to upper airway compromise, or from respiratory failure secondary to diaphragmatic and intercostal muscle paralysis.

Diagnosis of botulism is most often made by detection of the specific BoNT in food, vomitus, intestinal contents, or other bodily fluids by PCR and EIA or the definitive mouse neutralization assay. Testing is most often performed by public health laboratories rather than clinical microbiology laboratories. Culture of suspect foods, soil, and/or dust samples may also result in the isolation of *C. botulinum* or other BoNT-producing clostridia. Confirmation of culture identification generally requires a toxin neutralization assay, since there are many clostridial species that are phenotypically similar to *C. botulinum* that do not produce toxin (e.g., *C. sporogenes, C. baratii*).

Treatment of botulism requires intensive supportive care, including ventilator-assisted respiration for extended periods of time until the flaccid paralysis resolves. Specific antitoxin can neutralize circulating BoNT, lessening the effects of the toxin if given early, but does not neutralize toxin that is already bound to nerve tissue. Thus paralysis does not immediately subside with antitoxin therapy and continued supportive care is necessary.

TABLE 246.3 Characteristics of *Clostridium* sp. Producing Botulinum Neurotoxin (BoNT)

GROUP	DISEASE	TOXIN TYPES	SACCHAROLYTIC	PROTEOLYTIC	LIPASE
I	Humans, animals	A, B, F	Yes	Yes	Yes
II	Humans, animals	B, E, F	Yes	No	Yes
III	Birds	C, D	Yes	+/–	Yes
IV	Humans, animals	G	No	Yes	No

Clostridium tetani and Tetanus

Tetanus is caused by *C. tetani* and is most often associated with contamination and growth of *C. tetani* within puncture wounds, usually not considered as serious injuries. A potent neurotoxin is responsible for disease in the absence of neutralizing antibodies. In the United States, there are only about 50 cases of tetanus reported to the Centers for Disease Control and Prevention each year, because most people in Western countries are immunized with tetanus toxoid. Tetanus is a disease of nonimmunized people, usually in developing countries where vaccination against the neurotoxin is not widely practiced. The organism is found in the soil and intestinal contents of most animal species. Tetanus intoxication is similar to botulism and is caused by the neurotoxin tetanospasmin (TeNT). Unlike botulism, wherein the toxin does not affect the central nervous system, TeNT traverses the nerve terminal through the nerve body, with axonal transport to the spinal cord and brainstem. TeNT is synthesized as a single inactive polypeptide chain that is cleaved by a protease to produce an active di-chain consisting of a heavy chain (100 kDa) and a light chain (50 kDa) linked by a disulfide bond. The heavy chain facilitates binding to the neuron and the light chain enters the cell through endocytosis. The light chain is a zinc endopeptidase that cleaves the SNARE protein in the same place that BoNT acts, preventing exocytosis of specific neurotransmitters.[10]

The diagnosis of tetanus is usually made by clinical observation, since the organism is rarely cultured from infected sites. Confirmation of prior vaccination makes the diagnosis of tetanus extremely unlikely in vaccinated individuals. Treatment of tetanus generally occurs in intensive care settings with the administration of agents to control spasms, reduce anxiety, and promote muscle relaxation. Because pulmonary complications are common, respiratory support for extended periods of time is often necessary. Although rare in the United States and despite the availability of appropriate supportive care, the mortality from tetanus ranges from 25% to 50%, with pulmonary complications often as the cause of death. Recovery in uncomplicated cases takes 3 to 6 weeks.

Food Poisoning Caused by *Clostridium* Species

Clostridium perfringens type A causes virtually all cases of clostridial food poisoning throughout the world. Contaminated food products that are not properly cooked or stored allow for the proliferation of large numbers of vegetative *C. perfringens* cells. Once consumed, the vegetative cells sporulate within the small intestine, releasing an enterotoxin that causes the characteristic symptoms. The enterotoxin appears to be a cytotoxin in that it causes demonstrable damage to mammalian cells. The enterotoxin is a small polypeptide with a molecular weight of about 35 kDa. The toxin causes fluid accumulation in the rabbit ileal loop assay, provokes vomiting with orogastric challenge in experimental animals, and produces a characteristic cytopathic effect in Vero and other cell lines.[24,84] The *C. perfringens* enterotoxin gene *cpe* can be used to detect disease-causing strains. There is some evidence that the *cpe* gene can be carried on a plasmid and spread from one *C. perfringens* strain to another. More recently, the association between some cases of presumed AAC and *C. perfringens* enterotoxin have been reported.[25]

Clostridium perfringens food poisoning requires the ingestion of at least 10^8 viable enterotoxin-producing cells and occurs most often following the consumption of inadequately cooked and stored food. Meat and meat-containing foods are the most commonly implicated sources. Most cases occur as a common source outbreak, usually involving commercially prepared foods, and on average involve more than 20 affected individuals. Unlike *S. aureus* food poisoning and salmonellosis, home outbreaks appear to be uncommon. However, this may only reflect the fact that individual illnesses are not as likely to be identified as food poisoning when compared to group illnesses involving large numbers of people.

The incubation period for *C. perfringens* food poisoning is short, generally between 7 and 15 hours with a range of 6 to 24 hours. Symptoms include watery diarrhea, abdominal cramping, vomiting, and fever. Cases resolve spontaneously within 24 to 48 hours. When disease is suspected, culture of suspected food or stool from affected individuals is recommended, with quantitative determination of the number of *C. perfringens* present serving as a presumptive diagnostic indicator. If the isolated organisms are present at concentrations exceeding 10^6 cfu/g and the toxin gene is detected, *C. perfringens* type A is presumed to be the cause of the outbreak. Detection of the enterotoxin using a cytopathic toxin assay with neutralization can also be used as a diagnostic test. Some public health laboratories also employ latex agglutination or EIA for detecting the enterotoxin.

OTHER CLOSTRIDIAL INFECTIONS

Bacteremia

Although obligate anaerobes are infrequently isolated from bacteremic patients, clostridia are second only to *Bacteroides* among anaerobes isolated from blood cultures, accounting for approximately 1% of all positive cultures.[85,86] Blood culture data for a large Boston teaching hospital show that, over a 5-year period from 2003 to 2007, over 29,000 positive blood cultures were detected, with over 200 positive for obligate anaerobes (unpublished data). Of the positive cultures, 135 were positive for *Bacteroides fragilis* and 58 were positive for *Clostridium* spp. Of the *Clostridium* species isolated, *C. perfringens* accounted for 42 isolates, *C. septicum* for 12 isolates, and *C. difficile* for 4 isolates. Significant risk factors associated with isolation of clostridia from blood include hemodialysis, intestinal malignancy, and inflammatory bowel disease.[85] Patients undergoing treatments that render them neutropenic are also at increased risk for clostridial bacteremia; *C. perfringens,* followed by *C. septicum,* are the most common species isolated from blood. While isolation of clostridia from blood may reflect either a transient bacteremia due to infection at another site or contamination, isolation of certain species, such as *C. septicum,* may reflect the presence of an underlying intestinal malignancy (Fig. 246.4). If not treated promptly, the toxins produced by clostridia during septicemia can result in severe disease and devastating clinical outcomes.

Abdominal Infections

Infections resulting from contamination of the peritoneal cavity with intestinal contents are most often polymicrobial, and clostridia are often isolated from purulent material accompanying such infections, along with other obligate anaerobes and facultative species. Although a clear role in the infectious process has been established for organisms such as *B. fragilis* and the Enterobacteriaceae, the pathogenic significance of clostridia during these mixed infections is not clear. Gangrenous infections of the abdominal wall caused by *C. perfringens* have been well documented and usually are associated only with the presence of clostridia.

Biliary Tract Infections

Clostridia can be isolated from over 20% of diseased gallbladders and represent contamination of the biliary tract by normal intestinal bacteria. Biliary tract infections may either involve a single clostridial pathogen or reflect a mixed microbiota. *Clostridium perfringens* accounts for 50% of the clostridial species isolated. In some cases, gas may be present in the gallbladder lumen along with purulent material.[1] Gas gangrene of the abdominal wall is a rare complication of gallbladder surgery, resulting in contamination of the peritoneal cavity by the diseased biliary tract tissue. Demonstration of gas within the biliary tract by radiographic methods is an indication for emergent surgical intervention and prompt antibiotic treatment directed at both obligate anaerobes, including clostridia and facultative enteric organisms.

Female Genital Tract Infections

Clostridia can be isolated from the genital tract of approximately 10% of women as part of the normal vaginal microbiota. Clostridia are present in up to 20% of non–sexually transmitted disease genital infections and may be present as part of bacterial vaginosis.[87] Although not common, postpartum and postabortion infections caused by clostridia, particularly *C. perfringens* and *C. sordellii,* can be severe.[88,89] Clostridial uterine infections start as localized chorioamnionitis as a result of infection of the fetus and placental tissues. Often a gaseous vaginal discharge is present. The infection may spread to the uterine wall and endometrial

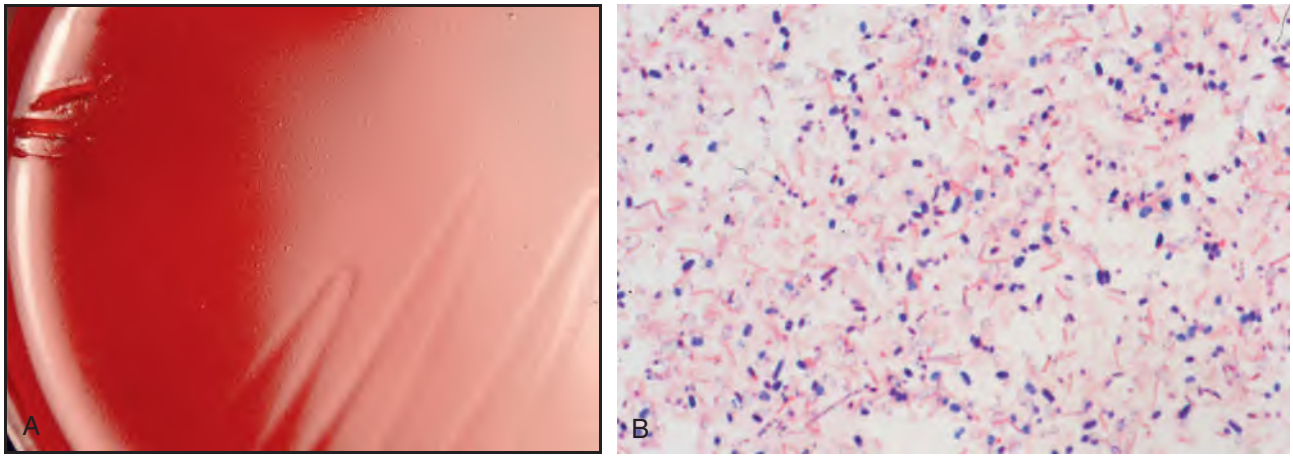

FIG. 246.4 ***Clostridium septicum.*** (A) *Clostridium septicum* growth on blood agar. Discrete colonies are not seen, and growth appears as a swarming film on the surface of the agar plate. (B) Gram stain of *C. septicum* demonstrating the gram-variable nature and the pleomorphic appearance of this species.

tissues, and in the most severe cases uterine necrosis accompanied by sepsis ensues.[1]

Pleuropulmonary Infections

Clostridia have been recovered from up to 10% of anaerobic pulmonary infections, with *C. perfringens* accounting for the majority of isolates.[1] Clostridia are rarely isolated as pure cultures from these specimens, suggesting that their presence as part of a mixed microbiota does not necessarily reflect a causative role in the disease process. The most common sources for clostridia in such infections include oral microbiota and aspirated stomach contents. The clinical features of infections from which clostridia can be isolated are similar to those for other obligate anaerobes. On rare occasions, gas within the pleural space can be detected in association with the presence of clostridia.

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The complete reference list is available online at Expert Consult.

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247 *Bacteroides, Prevotella, Porphyromonas,* and *Fusobacterium* Species (and Other Medically Important Anaerobic Gram-Negative Bacilli)

Wendy S. Garrett and Andrew B. Onderdonk

SHORT VIEW SUMMARY

Definition
- *Bacteroides, Porphyromonas, Prevotella,* and *Fusobacterium* spp. account for most infections caused by gram-negative anaerobic rods. *Bilophila* and *Sutterella* spp. also cause human infections, although they are less frequently encountered.

Epidemiology
- These organisms are part of the normal human microbiome and can be isolated from the oral cavity, gastrointestinal tract, and vaginal vault of humans.

Microbiology
- The organisms are obligately anaerobic, gram-negative, non–spore-forming rods. Members of this group can be proteolytic, saccharolytic, or both. Some species produce catalase and superoxide dismutase in low concentrations. Some species have capsular polysaccharides that have been shown to be potent immunomodulators, whereas other species produce abundant proteases that can degrade tissue.

Diagnosis
- Identification in the clinical laboratory includes colony morphology, Gram stain, pigment production visualized in natural light and as fluorescence emission after exposure to ultraviolet light, and numerous biochemical tests (e.g., Vitek). More recently the use of matrix-assisted laser desorption/ionization time-of-flight mass spectrometry proteomic identification methods have been employed for some species.

Therapy
- Treatment is directed by culture and sensitivity test results (see Table 247.2). In general, β-lactam antibiotics in conjunction with β-lactamase inhibitors show excellent activity against *Bacteroides fragilis* group members and *Prevotella, Porphyromonas, Fusobacterium, Bilophila,* and *Sutterella* spp. First-generation cephalosporins are ineffective against the *B. fragilis* group and *Prevotella* and *Porphyromonas* spp. Clindamycin is a highly effective antibiotic against *Prevotella, Porphyromonas,* and *Fusobacterium* spp.; however, there is significant resistance among the *B. fragilis* group and *Sutterella* spp.

OVERVIEW

The genera *Bacteroides, Porphyromonas, Prevotella,* and *Fusobacterium* account for most infections caused by gram-negative anaerobic rods (GNARs). *Bilophila* and *Sutterella* also cause human infections, although they are less frequently encountered in clinical practice. These obligately anaerobic gram-negative bacteria colonize the oropharynx, gastrointestinal tract, and urogenital tract of humans. Several species from some of these genera are useful symbiotic bacteria, facilitating host metabolism and favorably shaping immune responses. However, many of these microbes act opportunistically, causing infections when they gain access to otherwise sterile tissues. When tissue barriers are breached, GNARs have a predilection for abscess formation, with the most common sites being the oropharynx, abdominal cavity, lungs, and female genital tract. These bacterial species also present clinical challenges because they are often resistant to commonly used antibiotics. A few strains from the *Fusobacterium, Bilophila,* and *Sutterella* genera have been associated with either inflammatory bowel disease or colon cancer, although causative roles remain to be robustly established outside of preclinical models.

HISTORY

The end of the 19th century was a fertile time for the discovery of GNARs. The recognition of GNARs as important pathogens first occurred in animals in 1884 and is attributed to Loeffler. Schmorl is credited with identifying GNARs as opportunists in humans in 1891. Identification of GNARs as members of the human microbiota dates back to 1898, when Halle published his discovery of *Fusobacterium* colonizing the female genital tract. At the same time, GNARs were also isolated by Veillon and Zuber from clinical material from pelvic, appendiceal, and brain abscesses.[1]

The *Bacteroides* spp. as a group were first described in the late 1890s, and for many years *Bacteroides* was a physiologically disparate genus of pleomorphic, obligately anaerobic, gram-negative rods. Until the early 1960s, competing taxonomic systems and confusion regarding nomenclature made it difficult to determine the role of specific members of this group of organisms during infectious processes. Holdeman and colleagues[2] brought some conformity to the phenotypic classification of this group of organisms, which allowed other investigators to identify *Bacteroides fragilis* as a clinically significant abscess-inducing pathogen. With the advent of uniform taxonomic classification and the use of 16S ribosomal RNA phylogenetic-driven taxonomic classifications, several species were reclassified. *Bacteroides* has been further subdivided into two additional genera—*Porphyromonas* and *Prevotella*—both of which primarily colonize the oral cavity. The genus *Fusobacterium* was identified in the early 20th century and is well documented in studies by microbiologists at the Virginia Polytechnic Institute and Pasteur Institute.

MICROBIOLOGY

Bacteroides

Bacteroides is a genus of gram-negative, non–spore-forming, obligately anaerobic, rod-shaped bacteria. More than 30 species of *Bacteroides* have been recognized. The strictest taxonomic definition of *Bacteroides* limits this census to less than a dozen separate species. Within the Bacteroidaceae family, *Bacteroides* organisms are distinctive in their DNA guanine-cytosine composition—40 to 48 mol%. The principal products of their saccharolytic metabolism are acetate, succinate, and

isovalerate. The saturated anteiso-methyl and isomethyl branched-chain fatty acids are used for their long-chain fatty acid–based identification. *Bacteroides* organisms express hexose monophosphate shunt–pentose phosphate pathway enzymes. Their sphingolipid-rich membranes also possess menaquinones, particularly MK-10 and MK-11. *Bacteroides* peptidoglycan contains meso-diaminopimelic acid. Several *Bacteroides* spp. express numerous capsular polysaccharides. These glycoantigens are of interest biologically for their immunomodulatory potential, particularly in the case of *B. fragilis* polysaccharide A.[3]

GNARs are differentiated from one another in the clinical laboratory by use of standard techniques: colony morphology, Gram stain, pigment production visualized in natural light and as fluorescence emission after exposure to ultraviolet light, and numerous biochemical tests. Short-chain fatty acid analysis can also be used for species-level *Bacteroides* discrimination. The *B. fragilis* group is of special medical importance for several reasons. It is often the predominant GNAR in polymicrobial infections, and members of this group have the potential to express β-lactamase. *B. fragilis* group bacteremias are also associated with a high mortality rate of 27%.[4] Consequently, identification of the *B. fragilis* group by the clinical laboratory is often critical for appropriate therapeutic intervention. On blood agar, *B. fragilis* forms circular, entire, white or gray, 2- to 3-mm colonies that are shiny and smooth. The *B. fragilis* group can be pleomorphic on Gram stain, forming straight rods of varying length as well as coccobacilli (Fig. 247.1A). When grown in liquid medium, cells develop bipolar vacuoles and show a characteristic "safety pin" appearance. A useful characteristic of the *B. fragilis* group is bile tolerance compared with other GNARs, enabling its growth on *Bacteroides* bile-esculin agar. In addition, *B. fragilis* is highly resistant to the antibiotics kanamycin, vancomycin, and colistin. The use of a simple disk diffusion assay for these antibiotics is often part of the identification process for GNARs (Table 247.1).

Prevotella and *Porphyromonas*

Both *Prevotella* and *Porphyromonas* were previously considered to be part of the genus *Bacteroides*. These pigmented GNARs can be distinguished from one another metabolically as the saccharolytic *Prevotella* spp. and the asaccharolytic *Porphyromonas* spp. Approximately 20 *Prevotella* spp. have been implicated in causing human disease. *Prevotella* forms circular, convex, 1- to 2-mm, shiny, gray colonies. On Gram stain, they form short gram-negative rods and may assume coccobacilli forms (Fig. 247.1B). *Prevotella* grows well on laked blood agar with kanamycin and vancomycin (LKV) and has variable resistance to colistin. Although *Prevotella* spp. are largely regarded as pigmented GNARs, they can be nonpigmented as well. Pigmented *Prevotella* spp. form brown or black colonies after a week of growth on LKV. Before this brown or black pigment develops, *Prevotella* may fluoresce a dark red on exposure to a Wood lamp (long-wave ultraviolet light). By colony morphology and Gram stain, *Porphyromonas* spp. tend to form smaller colonies and appear as shorter rods or coccobacilli on Gram stain but can be difficult to distinguish from *Prevotella*. *Porphyromonas* usually grows as pigmented colonies, initially forming gray colonies that darken to black colonies within a week after plating on laked blood agar. *Porphyromonas* does not grow on LKV media because of its sensitivity to vancomycin, but it is resistant to colistin.

Fusobacterium

Fusobacterium is a genus of obligately anaerobic filamentous gram-negative rods that are members of the phylum Fusobacteria, in contrast to the *Bacteroides, Prevotella,* and *Porphyromonas* genera, which are members of the phylum Bacteroidetes. On blood agar, *Fusobacterium* forms pinpoint colonies that can be circular or irregular, with some species, such as *Fusobacterium nucleatum,* forming umbonate "fried egg" colonies after 3 to 5 days of incubation. Depending on the strain, they can be hemolytic, and some strains hemagglutinate. *Fusobacterium* spp. can be variable on Gram stain and display a range of cellular morphologies from coccoid, pleomorphic spherules *(Fusobacterium necrophorum)* to rod shaped. Rods can be short with rounded ends or long and thin with pointed ends *(F. nucleatum)*, arrayed end to end (Fig. 247.1C). As a genus, *Fusobacterium* is sensitive to both kanamycin and colistin and resistant to vancomycin. It can be distinguished by its bile sensitivity and metabolism of threonine to propionate. Most species are indole positive and produce butyric acid during the fermentation of glucose.

Symbiosis

The human microbiome is dominated by anaerobes. GNARs colonize the mucosal surfaces of the oropharynx, gastrointestinal tract, and female urogenital tract, and *Bacteroides* and *Prevotella* are among the most abundant genera present. Mutualism and opportunism are important features of the symbiotic relationship between human hosts and colonizing species of the *Bacteroides, Prevotella, Porphyromonas,* and *Fusobacterium* genera.

TABLE 247.1 Techniques and Properties to Differentiate Medically Important Gram-Negative Anaerobic Rods

	ANTIBIOTIC SENSITIVITY (PENCILLIN [P], VANCOMYCIN [V], COLISTIN [C], KANAMYCIN [K])	GROWTH IN 20% BILE	PIGMENT	BRICK RED FLUORESCENCE
Bacteroides fragilis group	$P^RV^RC^RK^R$	Y	N	N
Prevotella	$P^SV^RC^VK^R$	N	Y	V
Porphyromonas	$P^SV^SC^RK^R$	N	Y	V
Fusobacterium	$P^SV^RC^SK^S$	V	N	N

N, No; *R,* resistant; *S,* sensitive; *V,* variable; *Y,* yes.

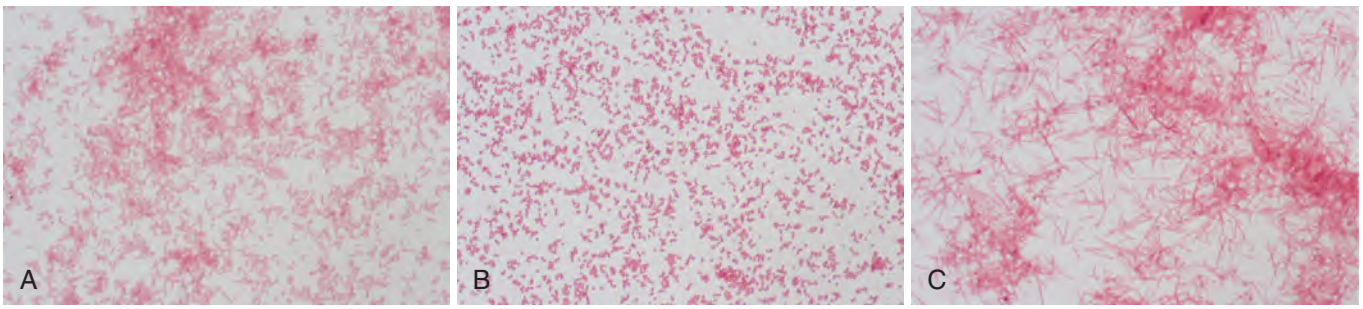

FIG. 247.1 Gram stains of selected gram-negative anaerobic rods. (A) *Bacteroides fragilis.* (B) *Prevotella intermedia.* (C) *Fusobacterium nucleatum.*

Gastrointestinal Tract

The human colon is colonized by 10 trillion to 100 trillion bacteria, making it the largest repository for bacteria in the body. The *Bacteroides* genus constitutes 30% of the total colonic bacteria. *Bacteroides vulgatus, Bacteroides thetaiotaomicron, B. fragilis,* and *Bacteroides ovatus* are the most commonly encountered *Bacteroides* spp. in the human colon.[5] There is significant variability among humans regarding the colonic colonization, with different *B. fragilis* group members as assessed by stool culture and 16S ribosomal RNA gene amplicon sequencing (Comstock and Onderdonk, unpublished). The colonization of the gastrointestinal tract occurs during descent through the vaginal canal or postnatally for infants delivered by cesarean section. In particular, children born by cesarean section appear to have a delay in reaching their full *Bacteroides* complement.[6] Both breast milk and exposure to environmental factors are important forces in shaping colonization. Many studies over the past several decades have been undertaken to examine this process in detail, with genomic-based technology taking center stage more recently.

SYMBIOSIS AND MUTUALISM IN IMMUNITY AND METABOLISM

There has been an explosion of interest in clinical correlations between microbiota profiles including the presence or absence of specific species and human health and disease. The pioneering work of Björkstén[7] and others examined correlations between infectious diseases and atopic and allergic diseases for many decades. One study suggested that *B. fragilis* colonization at younger than 3 weeks of age increases the risk of asthma later in life.[8] The basic science behind these clinical observations rests in the hypothesis that certain bacterial factors not only instruct immune cell development but also tune immune responsiveness. A large body of work in preclinical models over the past decade supports that the *B. fragilis* zwitterionic polysaccharide A (PSA) reinforces homeostatic immune function both in the gut and systemically despite earlier work that focused on the abscessogenic potential of *B. fragilis*. Investigators have demonstrated that PSA is a symbiosis factor that balances T-cell subset population size and function and may be a beneficial microbe for the treatment of chronic inflammatory diseases such as inflammatory bowel disease and multiple sclerosis.[9]

The immunomodulatory effects of *Bacteroides* spp. are not exclusive to *B. fragilis; B. thetaiotaomicron* also has immunomodulatory properties with the potential to dampen proinflammatory responses to commensal, intestinal bacteria. *B. thetaiotaomicron* targets the RelA subunit of the transcription factor nuclear factor kappa B, a master regulator of proinflammatory immune responses. This process is dependent on a nuclear receptor and transcription factor called *peroxisome proliferator-activated receptor gamma* and is independent of the nuclear export receptor CRM1.[10] However, the mechanism by which *B. thetaiotaomicron* drives this process remains to be fully understood. Strains of *B. thetaiotaomicron* with sulfatase activity have also been shown to contribute to intestinal inflammation in genetically susceptible mice.[11,12] *Bacteroides* and other intestinal commensals also contribute to host immunity through the process of colonization resistance, the concept that the entrenched presence of gut symbionts provides protection from invading pathogens. The most central and well-understood function of *Bacteroides* spp. involves the mutualistic function of metabolism. The human gut at its core is a bioreactor. The saccharolytic *Bacteroides* spp. process diverse dietary and host polysaccharides for their own metabolic needs and in doing so aid in human digestion and nutrient liberation for their human hosts. A well-nuanced understanding of the biochemistry and genomics of glycan foraging by *B. thetaiotaomicron* has emerged over the past several years. In addition, numerous studies support the potential role of these bacteria in human energy balance with implications for the human obesity epidemic.[13]

Female Urogenital Tract

The female urogenital tract, particularly the vagina, is densely colonized by anaerobes. Although gram-positive anaerobic lactobacilli are the predominant colonizers, GNARs may also be present in significant numbers. Vaginal colonization is dynamic, with variations not only intrinsic to the female reproductive life cycle from the premenarchal through postmenopausal years but also with significant shift over a given menstrual cycle.[14] Pregnancy and parturition result in substantial microbiota population shifts in both the urogenital tract and the gastrointestinal tract as well.[15,16]

There is also intraindividual genus-level diversity in colonization with *Bacteroides, Fusobacterium, Prevotella,* and *Porphyromonas* genera. In addition, distinct microbial community differences exist among the labia, the urethral vestibular region, the length of the vagina, and the cervix. Frequent isolates from the vaginal vault include *Bacteroides urealyticus* and members of the *B. fragilis* group; *Prevotella bivia, Prevotella disiens, Prevotella buccalis, Prevotella melaninogenica,* and *Prevotella corporis; Porphyromonas asaccharolytica;* and *F. nucleatum.* Most research on the vaginal microbiota and symbiotic effects has focused on *Lactobacillus,* with the view that GNARs may contribute to vaginal dysbiosis, negative consequences for fetal outcomes, and host mycotic and bacterial infections.

Oropharynx

The oropharynx is a diverse niche for both aerobes and anaerobes. Within the mouth, the teeth, gingival crevices, saliva, and posterior pharyngeal structures provide distinctive milieus with diverse pH and oxygen tensions. Gingival scrapings are particularly dense in bacteria, with estimated concentrations on the order of 10^{11-12} CFU/mL. Similar to the colonization of the lower gastrointestinal tract, colonization of the oropharynx starts at birth. Among the anaerobes, *Lactobacillus* and *Peptostreptococcus* are the earliest colonizers. Fusobacterial populations emerge with the eruption of the first teeth and increase with establishment of full juvenile dentition. Colonization with *Prevotella* and *Porphyromonas* spp. emerges after colonization by *Fusobacterium* spp. Of the GNARs, *Porphyromonas gingivalis* and *P. endodontalis; F. nucleatum; Prevotella intermedia, P. melaninogenica, Prevotella denticola,* and *Prevotella loescheii;* and *Tannerella forsythia* all commonly populate dental plaque. Poor dentition, gingivitis, and other periodontal diseases correlate with increased numbers of GNARs, as do hospitalization, residence in a long-term care facility, and multiple medical comorbidities.

Opportunism

Symbiosis should be viewed as a host-microbe relationship spanning the spectrum from mutualism through opportunism. Most, if not all, symbiotic GNARs have pathogenic potential. *B. fragilis* warrants special consideration because although *B. fragilis* itself may not be the most abundant GNAR cultured from the gastrointestinal tract, it is the most common GNAR identified in clinical isolates from both blood and abscesses.[17] Multiple virulence factors underpin this observation and account for the observed opportunism of *Prevotella, Porphyromonas, Fusobacterium,* and *Bilophila.* Virulence factors produced by these bacteria include capsular polysaccharides, outer membrane proteins, lipopolysaccharide (LPS) endotoxins, attachment factors (fimbriae/pili/adhesins), toxins, and numerous enzymes. Synergism is another important concept that explains the presence of GNARs in many polymicrobial infections. Several facultative anaerobes and aerobes can provide favorable environments that can promote the growth of GNAR. For example, the production of superoxide dismutase by facultative species protects obligate anaerobes that do not produce this enzyme constitutively against the highly lethal superoxide anion. Without the presence of these other microbes, the oxygen tension would not be favorable for GNARs. In many polymicrobial infections, facultative anaerobes and aerobes function mutualistically for GNARs, providing strong synergy for their expansion, especially for *B. fragilis.* In turn, the *B. fragilis* group with its frequent β-lactamase activity can provide a protective environment for normally β-lactam-sensitive facultative anaerobes and aerobes. Such cooperativity can also translate into an interdependency that may explain why antimicrobial therapy that is not effective against every member of a polymicrobial infection is nevertheless able to successfully cure some infections.

Endotoxic Lipopolysaccharide

Endotoxic LPS is a powerful mediator of systemic inflammation and a driver of septic shock. LPS can differ in its endotoxic potential. Because of the structure of lipid A in *Bacteroides,* it lacks C14-2OH fatty acid,

and the LPS does not elicit a strong host inflammatory response. However, LPS immunogenicity of gut microbiome GNARs such as *Bacteroides* spp. may be of considerably more biologic significance than previously appreciated, as levels have been associated with susceptibility to autoimmune diseases including type 1 diabetes in human populations.[18] *Porphyromonas* and *Prevotella* spp. appear to have a similarly attenuated LPS. *P. gingivalis* LPS may be able to antagonize the proinflammatory effects of other LPS in mixed infections, as suggested by in vitro experiments on primary human monocytes. In contrast, *Fusobacterium* LPS elicits a more inflammatory host response because its lipid A contains C14-2OH fatty acid.

Capsular Polysaccharides

B. fragilis produces eight distinct polysaccharides, PSA to PSH, and there is complex phase variation in the expression of these polysaccharides. There is also substantial variability in these polysaccharides across *B. fragilis* strains.[19] Over the past 30 years, the work of Kasper, Onderdonk, Comstock, and Mazmanian has led to a nuanced understanding of the role of these polysaccharides in abscess formation and immunomodulation. The capsular polysaccharide complex facilitates binding of *B. fragilis* to mesothelial cells lining the peritoneum providing a nidus for abscess formation and in the intestinal mucosa helps to reinforce balanced T-cell immune responses. Numerous *Porphyromonas* and *Prevotella* spp. including *Prevotella oralis* and *P. melaninogenica* as well as *Fusobacterium* spp. are also encapsulated and are abscessogenic.

Pili and Fimbriae

In addition to capsular polysaccharide complexes promoting adherence, *Bacteroides* and *Porphyromonas* spp. can adhere to the epithelium via pili. There are great variations in cellular adherence of different *Bacteroides* spp. and strains. Pili are more frequently observed in pathogenic clinical isolates than in fecal samples from healthy subjects.[20] Pili have been observed specifically in *B. fragilis* and *B. ovatus* strains as well as in *P. gingivalis* and *F. necrophorum,* and other *Fusobacterium* spp. express numerous adhesins.

Enzymes and Toxins

Numerous enzymes and toxins facilitate GNAR host mucosa invasion and immune system evasion. *Bacteroides, Prevotella, Porphyromonas,* and *Fusobacterium* spp. can also express numerous extracellular enzymes (collagenase, chondroitin sulfatase, hyaluronidase, neuraminidase, deoxyribonuclease, phosphorylase, and proteases) that can facilitate epithelial barrier breach. Enterotoxigenic strains of *B. fragilis* (enterotoxigenic *B. fragilis* [ETBF]) cause diarrhea in both children and adults.[21] The toxin Btf is a 20-kDa zinc-containing metalloprotease, and there are at least three isoforms. Btf is posited to cleave e-cadherin in the zonula adherens, resulting in intestinal barrier compromise.

Metabolic End Products

Numerous *Bacteroides* and *Prevotella* spp. produce significant amounts of succinate as an end product of metabolism. Several studies have suggested that succinate inhibits polymorphonuclear leukocyte (PMN) phagocytosis. In addition, a succinate-rich environment may inhibit PMN migration and perhaps drive PMN apoptosis.

Host Immune Response

Cellular and noncellular innate and adaptive immune responses play important roles in defending the host from infections from the GNAR. Neutrophils are an important first line of defense. Phagocytic and oxygen radical–generating abilities are not the only defense of the neutrophil against pathogenic GNARs. Neutrophils also produce a number of antimicrobial peptides that are bactericidal for GNARs. GNAR-positive abscesses are often neutrophil rich, and neutrophils can kill *Bacteroides* spp. and other GNARs under anaerobic conditions. Further support for the role of neutrophils in protecting the host from GNAR infections comes from studies of the prevalence of gram-negative bacteremia in neutropenic patients. The complement system is another innate immune mechanism important in the clearance of these infections. Both the classical and the alternative complement pathways opsonize several *Bacteroides* and *Prevotella* spp. Most discussions of dendritic cells and *Bacteroides* spp. have not revolved around host defense but instead have focused on how dendritic cell–internalized glycoantigens can direct T-cell responses. In mouse models, immunization with PSA has been shown to protect from *B. fragilis*–induced abscesses. The protective effects of immunization were shown to be dependent on $CD4^+$ T cells, despite the fact that polysaccharides are regarded as T-cell–independent antigens. Furthermore, recombinant interleukin-2, a potent cytokine for T-cell proliferation, was protective against *B. fragilis* abscess formation in a dose-dependent manner, further substantiating a role for T cells in host-adaptive immunity to *Bacteroides* spp. B-cell deficiency has been shown to be a risk factor in mice for disseminated anaerobic infections, and passive immunity has been shown to be protective in mouse models. In rat models, passive transfer has not been shown to be protective. In general, there is little direct clinical evidence linking B-lymphocyte and T-lymphocyte function to host immunity to GNARs.

INFECTIONS

On primary evaluation few initial signs and symptoms implicate GNARs specifically. The clinical signs and symptoms of GNAR infection depend on the location of the infected site. A careful history with consideration of potential sites of translocation or portal of entry may raise the suspicion for involvement of GNARs (e.g., bacteremia in a patient with an intraabdominal process). Systemic infections such as bacteremia involving GNARs are often polymicrobial. Most of these infections arise endogenously from GNARs already colonizing the mucosal surfaces. Common clinical signs and symptoms associated with anaerobic infections in general include physical proximity to a mucosal site, foul-smelling purulent discharge, tissue necrosis, and palpable or imaged gas in tissues. In the present age of molecular diagnostics, clinicians should remember to engage their senses and in particular their noses in patient evaluations, as anaerobic empyema and peritonitis can often be diagnosed by smell at the bedside quickly and at no cost.

Bacteremia

The mortality of GNAR bacteremia has been reported to range from 15% to 60%, with the upper range of this mortality reflecting untreated infections.[22] In a retrospective, multicenter study from France, anaerobes accounted for 0.5% to 9% of all bacteremias, of which 60% were *Bacteroides* spp. and 22% were *Clostridium* spp. These figures are consistent with prior data over the past several decades.[23] In the United States, the estimated prevalence of GNAR bacteremia over the past 3 decades has fluctuated between 0.5% and 15%. Until more recently, the data suggested a trend toward a decreased incidence. A study from the Mayo Clinic reported that, at this large center at least, cases of anaerobic bacteremia are on the rise, with 53 cases per year reported during the years 1993–96, 75 cases per year reported during the years 1997–2000, and 91 cases per year reported during the years 2001–04.[24] During the period 2008–12 in one Boston hospital, approximately 55,000 blood cultures were performed per year, and approximately 10% were positive for growth; 0.08% of these were positive for GNARs, and 0.05% were *B. fragilis* cases (Onderdonk, unpublished observation). Because of the lack of specific signs and symptoms of GNAR bacteremia, clinicians are highly dependent on the clinical laboratory for diagnosis. At most US hospitals, blood samples from potentially bacteremic patients are routinely collected into culture systems, allowing the evaluation of anaerobes. There should be a low clinical threshold for broad empirical coverage in critically ill patients with histories consistent with GNAR bacteremic sources.

Of all the GNARs, *B. fragilis* is most commonly isolated in bacteremias, and there is variation in mortality rates reported among *B. fragilis* group members. Mortality rates of 24% and 31% have been associated with *B. fragilis,* rates of 50% have been associated with *Parabacteroides distasonis,* and rates of 100% have been associated with *B. thetaiotaomicron.*[25,26] Rapid susceptibility testing and administration of appropriate antibiotic therapy are essential for optimal clinical outcomes, as substantiated by an observational study of *Bacteroides* bacteremia that demonstrated 16% mortality with optimal therapy and 45% mortality without.[27] *Fusobacterium* bacteremias represent less than 1% of all bacteremias and less than 10% of anaerobic bacteremias. Of the *Fusobacterium* spp. causing bacteremia, *F. nucleatum* and *Fusobacterium mortiferum* are

the most common. Subclinical and transient bacteremia with *Prevotella* and *Fusobacterium* spp. has been documented after dental cleaning. Clinically significant bacteremia with *Prevotella* spp. has been reported after both periodontal and obstetric procedures. GNAR bacteremias may occur as secondary infections in the setting of other primary GNAR infections. Intraabdominal infections are the most frequent source of these bacteremias. Frequent intraabdominal processes associated with GNAR bacteremia include abscesses, surgical procedures, intestinal perforation, obstruction, colorectal cancer, and other malignancies. The upper or lower respiratory tract is often the primary site for fusobacterial bacteremia. The female genital tract, oropharynx, skin ulcers, and skeletal and soft tissues should be thoroughly investigated as sources of bacteremia because they are the associated source in 5% to 25% of cases.

Skeletal Infection

Gram-negative anaerobes can infect both joints and bones, as reviewed in depth by Brook.[28] Septic arthritis caused by GNARs generally results from either a hematogenous seeding of the joint space or direct extension of the bacteria via the skin. The large lower extremity joints—hips and knees—are the most common sites, followed by the shoulders and elbows. Surgery, trauma, and multiple medical comorbidities all are risk factors. Most cases are associated with single species rather than a polymicrobial infection, as is often seen with abscesses. In a classic review of both pediatric and adult cases of septic arthritis, Finegold[29] reported on more than 1200 anaerobic joint infections and found that *F. necrophorum* was the most common isolate recovered.[30] *B. fragilis* group bacteria can also cause septic arthritis, and these infections are often secondary and distant from the primary site. Anaerobic osteomyelitis tends to be polymicrobial. *Bacteroides* and *Fusobacterium* spp. are most frequently cultured. Not surprisingly, *Prevotella* and *Porphyromonas* spp. have been recovered from osteomyelitis cases involving bites. Mastoid bone infections often involve these oropharyngeal commensals as well. Patients with sickle cell disease, who are at increased risk for osteomyelitis secondary to sickle-related bony infarction, are the subject of several case reports of GNAR osteomyelitis involving both the axial and the appendicular skeleton.[31] Patients with diabetes and peripheral vascular disease are another risk group for anaerobic osteomyelitis; these infections often develop in the setting of chronic foot ulcers.

Skin and Soft Tissue Infection

GNARs are important pathogens in surgical wound infections, bites, ulcers, and infected pilonidal cysts. Following intraabdominal or gynecologic surgery, wounds can become infected with *Bacteroides* and *Prevotella* spp., resulting in infections of proximal skin and soft tissues. Human, dog, and cat bites can result in GNAR skin infections. *Prevotella* is a common anaerobe that infects dog bite wounds. With routine availability of anaerobic culture and non–culture-based identification methods, anaerobes are increasingly being recovered and detected from infected human and animal bites, especially bites complicated by abscesses. Pilonidal cysts and sacral decubitus ulcers can readily become contaminated by feces and subsequently infected by GNARs. Similarly, cutaneous abscesses, carbuncles, and furuncles in the perineal region can become infected with GNARs. *B. fragilis* is frequently cultured from decubitus ulcers in both elderly and pediatric populations as well as from foot ulcers from patients with peripheral vascular disease or diabetes.

Central Nervous System Infection

The most common central nervous system (CNS) infection is meningitis; however, anaerobic meningitis is extremely rare. In Finegold's historical analysis of anaerobic infections,[29] 298 cases of meningitis were reviewed. However, more than half were without substantiated culture data. In a later review of pediatric populations by Law and Aronoff,[32] 271 cases were reviewed, of which the vast majority—more than 85%—were in the setting of brain abscesses. Anaerobic culture of cerebrospinal fluid is not routinely performed, and given the rarity of these infections, there should be a compelling reason to do so. Of the GNARs reported, *B. fragilis* and *F. necrophorum* are the most commonly isolated. In several of these cases, the upper respiratory tract or intestinal tract was the primary source that resulted in hematogenous spread in patients with medical comorbidities compromising the integrity of the blood-brain barrier. Chronic and acute otitis media have also been implicated in several of these rare cases.

Anaerobic infections of the CNS typically manifest as abscesses and are frequently polymicrobial. When abscesses develop outside of the brain parenchyma and around the dura mater, they are referred to as *subdural empyemas* or *epidural abscesses,* depending on the location. The location of the brain abscess correlates with the source of the infecting organism, often arising in adjacent structures (e.g., frontal lobe abscess and sinus infections, temporal lobe abscess, mastoiditis). A brain abscess stemming from bacteremia in the absence of focal trauma can arise throughout the lobes as a focal or multifocal process. Recent neurosurgery, trauma, and the presence of ventricular shunts are risk factors for GNAR abscesses. The site of primary infection not only informs abscess location but also narrows the spectrum of causative organisms. *Bacteroides, Fusobacterium, Prevotella,* and *Porphyromonas* spp. all have been isolated from brain abscesses.

Infections of the Aerodigestive Tract

Oropharyngeal Infections

GNARs are frequent culprits in infections of the teeth and peridontium. Most odontogenic infections arise in the setting of dental caries. Once opportunists have established themselves in a dental plaque, they can cause local infections or disseminate and seed locoregional sites via extension or distant sites via hematogenous spread. Extension to and infection of the sublingual, submandibular, and perimandibular spaces can result in Ludwig angina, a rapidly progressing infection of the floor of the mouth that without rapid surgical intervention can lead to death by asphyxiation. Although *Actinomyces israelii* is the most commonly identified microorganism, among the GNARs, *Fusobacterium* has been commonly isolated. Tongue piercing, a popular trend, also increases the risk of Ludwig angina.[33,34] Locoregional spread can result in infection of the maxillary sinus, cavernous sinus, or brain parenchyma. Severe facial cellulitis, such as periorbital cellulitis, is another complication of anaerobic dental caries infection. More distal sites of dental infections involving hematogenous spread include endocarditis, mediastinal or pleuropulmonary abscesses, or orthopedic infections.

Poor dental hygiene results in gingivitis, which can lead to more severe periodontal disease. Acute necrotizing ulcerative gingivitis, or Vincent angina, is the most severe manifestation of gingivitis. *F. necrophorum, P. melaninogenica, P. intermedia, F. nucleatum,* and *P. gingivalis* all have been implicated as causative agents in acute necrotizing ulcerative gingivitis as well as odontogenic infections. *B. ureolyticus* and *T. forsythia,* but not the *B. fragilis* group, have also been identified as causative agents in oral infections.

GNARs can result in peritonsillar abscess formation. Both *P. melaninogenica* and *F. necrophorum* are frequent GNAR isolates. A study of patients with sore throats presenting to a university clinic reported detection of *F. necrophorum* in 20.5% of patients and 9.4% of asymptomatic students.[35] An accompanying editorial emphasized the challenge in interpretation of these data.[36] One feared complication of peritonsillar abscesses that involves *F. necrophorum* is Lemierre syndrome—jugular vein septic thrombophlebitis. These septic emboli can seed the lungs and result in multiple systemic abscesses. Although infection begins in the throat, symptoms of pharyngitis are often resolving when symptoms and signs of Lemierre syndrome develop. Tenderness in the anterior cervical triangle is often present. Bacteremia is universally present. Septic emboli may cavitate or cause empyema. Contrast-enhanced computed tomography is useful in showing jugular vein thrombosis. Clinical presentation of sore throat in conjunction with pneumonia, lateral neck pain, or symptoms of septicemia should raise suspicion for Lemierre syndrome in children and adults in good health. Although cases of Lemierre syndrome seemed to diminish with the introduction of antibiotics for streptococcoal pharyngitis starting in the 1940s, after the 1990s the reported incidence started to increase for unclear reasons. Whereas 4 to 6 weeks of appropriate antibiotic coverage is a cornerstone of treatment, the use of anticoagulation has remained controversial with a paucity of evidence-based data to support its use.[37]

GNARs play a role in chronic sinusitis and acute exacerbations of chronic sinusitis but not in acute sinusitis, which in the absence of a

viral etiology is caused by *Streptococcus pneumoniae, Haemophilus influenzae,* and *Moraxella catarrhalis.* The predominant anaerobes recovered in chronic sinus infections are *Prevotella, Porphyromonas, Fusobacterium,* and *Peptostreptococcus spp*. Rare complications of chronic sinusitis include CNS abscesses.

Salivary Gland Infections

Infection of the salivary glands, usually the parotid glands, can result from viral or bacterial pathogens. *Staphylococcus aureus* is the most frequent organism associated with acute suppurative parotitis, and mumps virus can be a cause of acute parotitis. GNARs are the predominant anaerobes implicated in non–*S. aureus* parotitis.[38] *Prevotella, Porphyromonas,* and *Fusobacterium* spp. all have been reported.

Ear Infections

Aerobes, *S. pneumoniae* and *H. influenzae*, are the predominant causative organisms in bacterial acute and chronic otitis media in the pediatric population. Anaerobes have been found in serous effusions and transmeatal biopsy specimens from patients with chronic otitis media and acute exacerbations in the setting of chronic otitis media. In a study by Brook and Finegold,[39] culturing serous effusions from 114 patients with otitis media yielded data from approximately 40% of samples; aerobes predominated over polymicrobial anaerobic and aerobic populations, followed by single anaerobic isolates in 15%. *Prevotella* spp. were the second most commonly cultured anaerobes. Le Monnier and colleagues[40] published a retrospective study of their institution's experience with 25 pediatric acute otitis media cases caused by *F. necrophorum*. Of these patients, 44% had uncomplicated otitis media, 40% had acute mastoiditis, and 16% (4 patients) had Lemierre syndrome. In the classic study of the microbiology of chronic otitis media by Brook and Finegold,[39] the *B. fragilis* group and *P. melaninogenica* were the predominant anaerobes cultured.

Thoracic Infections

Anaerobic infections of the lung parenchyma and pleural space are relatively common. More specifically, these clinical infections include community-acquired and nosocomial pneumonias, lung abscesses, and pleural empyemas. Anaerobes can also result in acute mediastinitis in the setting of severe oropharyngeal infections or perforations in the upper gastrointestinal tract. Poor dentition, gingivitis, chronic obstructive pulmonary disease, cystic fibrosis, and neuromuscular diseases all are medical comorbidities that increase the risk of anaerobic pleuropulmonary infections. Smoking, alcoholism, conditions associated with impaired consciousness, and the inability to clear oral secretions (seizure disorder, dementia, severe cerebrovascular disease) all increase the risk of aspiration, which is a key inciting event in these pneumonias and empyemas. Obtaining good-quality sputum samples, those not contaminated with saliva, can be a clinical challenge that confounds identification of the causative organisms in these pneumonias.

Pleuropulmonary infections linked with aspiration events are commonly polymicrobial with both aerobic and anaerobic isolates. Viridans-group streptococci members are frequently cultured aerobes in these infections. Regarding the GNARs, *Prevotella buccae, P. disiens, P. intermedia,* and *P. melaninogenica; B. urealyticus* and *T. forsythia;* and *F. nucleatum* all have been associated with these infections. Notably, mixed streptococcal and anaerobic pleural infectious processes have a lower associated mortality than staphylococcal, enterobacterial, or polymicrobial aerobic infections.[41] Sputum of patients with cystic fibrosis is a favorable niche for many bacteria in the absence of overt infection. *Prevotella* spp. have been cultured in relatively high numbers from sputum of patients with cystic fibrosis, and *Pseudomonas aeruginosa* colonization correlates with increased anaerobic colonization.[42]

Cardiovascular Infections

Although GNARs are relatively uncommon causes of endocarditis and pericarditis, they are clinically important because of their antibiotic resistance and high associated mortality of 21% to 43%.[43] Of the GNARs, *B. fragilis* group bacteria are the most common causative agent for endocarditis. However, *Fusobacterium* spp., specifically *F. necrophorum,* and other *Bacteroides* spp. have been cultured in endocarditis. Primary infectious sources include the gastrointestinal tract, head and neck, and genitourinary tract, with hematogenous spread to the cardiac valves. Anaerobic endocarditis is similar to aerobic endocarditis in terms of its valvular pattern, male predominance, and risk factors (e.g., intravenous drug use). However, a prior clinical history of heart disease is not as common in GNAR endocarditis, and thromboembolic complications are more common than in aerobic endocarditis. Specifically, *B. fragilis* is associated with large valvular vegetations and subsequent diffuse thrombolic phenomena. *B. fragilis* expression of heparinase and other fibrinolytic enzymes may underpin these findings.[44] In addition to the classic thromboembolic phenomena of endocarditis, temporal lobe and renal emboli and portal vein thrombosis have been observed. *B. fragilis* group and *Fusobacterium* spp. are the GNARs associated with pericarditis. Cardiac surgery, trauma, gastrointestinal fistulas and perforations, and concomitant pleuropulmonary infections all are risk factors.

Intraabdominal Infections

Intraabdominal abscesses can occur after frank perforation stemming from a trauma, surgical procedures of the intestine or biliary tract, or intestinal cancer. Abscesses also form in the setting of inflammatory or infectious processes such as appendicitis, inflammatory bowel disease, diverticulitis, cholecystitis, or pancreatitis. *B. fragilis* is the prototypic anaerobe associated with intraabdominal abscesses. *Escherichia coli* is also a common isolate. The facultative anaerobe *E. coli* and *B. fragilis* can act synergistically, and both are often isolated from intraabdominal abscesses. The host response to the capsular polysaccharides of *B. fragilis* results in abscess formation. Studies in mouse models using intraperitoneal injection of *B. fragilis* have provided valuable insight into how adaptive and innate immune cell subsets as well as mesothelial cells contribute to intraabdominal abscess formation. It remains a scientific curiosity why *B. fragilis,* which makes up less than 0.5% of the intestinal microbiota, is the cause of the vast majority of intraabdominal abscesses.

Peritonitis

Patients with end-stage renal disease on long-term peritoneal dialysis are at increased risk for peritonitis, a cause of great morbidity and mortality in these patients. More than 80% of peritoneal samples in such cases of peritonitis are culture positive, and of these only 2.5% are associated with anaerobic organisms.[45] Peritonitis secondary to trauma such as gunshot wounds or perforation in the setting of intraabdominal surgery or an intraabdominal inflammatory process is more likely to involve a GNAR, especially the *B. fragilis* group. In perforation peritonitis the GNARs most frequently cultured are *B. fragilis, Fusobacterium* spp., *Prevotella* spp., *Porphyromonas* spp., and *Bilophila wadsworthia.*[46] *B. wadsworthia* is frequently isolated in peritoneal inflammation and abscesses arising in the setting of appendiceal perforation or gangrene. In neonates, peritonitis and abscesses most often occur in the setting of necrotizing enterocolitis.

Enteritis

ETBF has been implicated in enteritis, in acute diarrhea in children and adults, and as an inciting agent in inflammatory bowel disease. Diarrhea in these cases is severe, nonhemorrhagic, and accompanied by marked abdominal pain. Observational studies have reported on the constellation of symptoms and prevalence of ETBF in adult and pediatric populations and hospital-acquired diarrhea.[21] Intriguing links are also emerging between ETBF and human inflammatory bowel disease and colorectal cancer.[47]

Urogenital Tract Infections

GNARs play a central role in numerous non–sexually transmitted diseases of the urogenital tract. The bulk of these infections involve the female reproductive organs and include bacterial vaginosis, Bartholin cyst abscess, pelvic inflammatory disease, tubo-ovarian abscess, endometritis, chorioamnionitis, and wound infections secondary to gynecologic or obstetric procedures. Bacterial vaginosis is common with vaginal microbiota blooms of *Gardnerella vaginalis, Bacteroides* spp., *Prevotella* spp., *Mobiluncus* spp., and genital mycoplasmas. The instigating events of bacterial vaginosis—in particular the relative importance of anaerobes versus *G. vaginalis* in disease causation—remain controversial (see

Chapter 108). Bacterial vaginosis is a risk factor for preterm labor; as such, GNAR colonization of the vagina and reproductive structures may pose a risk for adverse pregnancy outcomes. GNARs also play a pathogenic role in acute and chronic prostatitis, prostatic and scrotal abscesses, and scrotal gangrene (Fournier gangrene). In these male urogenital infections, *B. fragilis, Prevotella* spp., and *Porphyromonas* spp. have been isolated, and antibiotic therapy directed at these organisms is part of successful treatment regimens.

RECENT ASSOCIATIONS OF GRAM-NEGATIVE ANAEROBIC RODS AND DISEASE

For several GNARs, metagenomics technology is finding or disproving associations with diseases. Numerous studies now support an association between *F. nucleatum* and colorectal cancer using human colorectal tissue metagenomics and metatranscriptomics data.[48,49] Follow-up studies are yielding insight into the potential mechanisms of causality of *F. nucleatum* in colorectal carcinogenesis.[50–53] In contrast, potential associations between *Sutterella wadsworthensis* and inflammatory bowel disease have not been proven with metagenomics or metatranscriptomics.[54] *B. wadsworthia* has garnered interest in inflammatory bowel disease pathogenesis on the basis of a study investigating links between a high-fat diet and experimental colitis using a mouse model.[55] The explosion of interest in the microbiome and its members is clearly creating a resurgence of research in GNARs.

THERAPY

Although antimicrobials are the mainstay of treatment for GNAR infections, treatment regimens may also involve interventional and surgical approaches as well as adjunctive therapies to facilitate healing and recovery. Selection of empirical antimicrobial therapy for infections involving GNARs is guided by a few general principles. Intraabdominal infections should include coverage for both anaerobes and coliform bacteria; either two-drug regimens or single agents are appropriate (specific antibiotics are discussed later). Urogenital tract infections are usually polymicrobial, involving coliforms, anaerobes, and streptococci, and broad-spectrum monotherapy or two drugs are appropriate. Similarly, skin and soft tissue infections are polymicrobial, involve both aerobes and anaerobes, and require broad coverage. CNS infections, particularly brain abscesses, should be treated with metronidazole, which has good CNS penetration, and a β-lactam–β-lactamase combination or a third-generation cephalosporin for streptococcal coverage. In addition to combating infection, antibiotics have a clear prophylactic role in surgery (see Chapter 318). Especially in the case of colorectal surgery, prophylactic antibiotics improve patient outcomes by reducing postoperative infections.[56] Coverage for both aerobes and anaerobes has led to a significant decrease in surgical wound infections.

Surgical Treatment

Surgical treatment is an essential modality for many GNAR infections. Incision and drainage are usually necessary for treatment of abscesses. Because necrosis is often a feature of infections complicated by GNAR, débridement of necrotic tissues is necessary for resolution of infections. Interventional radiologists frequently use fluoroscopy, ultrasound, or computed tomography guidance to place percutaneous drains that effectively drain abscesses. In many cases, surgical drainage is performed if relatively less invasive measures are unsuccessful. Location, size, and attendant procedural risk all are prominent factors in clinical management of abscesses.

Antibiotic Therapy

Treatment of many GNAR infections is empirical because of the polymicrobial nature of these infections and the time delays associated with anaerobic culture, identification, and susceptibility testing. Antibiotics that provide coverage across the aerobic and anaerobic spectrum should be administered when there is clinical suspicion of mixed infections. Many GNARs are resistant to a number of antimicrobials as a result of extended β-lactamase resistance and metronidazole resistance, making these infections a clinical challenge (Table 247.2).

For decades, penicillin G was the drug of choice for numerous GNAR infections. The *B. fragilis* group is penicillin resistant and, as such, penicillin G is used for GNAR infection outside of the abdominopelvic cavity. However, penicillin treatment failure has emerged secondary to β-lactamase production by certain GNARs. Penicillin resistance has been reported for *P. bivia* and *P. disiens, Porphyromonas* spp., *Bacteroides splanchnicus,* and *B. wadsworthia.* β-Lactamase-producing strains of *F. nucleatum,* both intraoral and extraoral, have been reported, and *F. necrophorum* is often penicillin resistant.[57] Although penicillin G at one time was considered a reasonable choice for minor odontogenic infections, it is not recommended as empirical coverage for severe oropharyngeal or pleuropulmonary GNAR infections because of concerns about resistance.

β-Lactam antibiotics in conjunction with β-lactamase inhibitors (e.g., ticarcillin-clavulanate or piperacillin-tazobactam) show excellent activity against *B. fragilis* group members as well as *Prevotella*, *Porphyromonas, Fusobacterium, Bilophila,* and *Sutterella* spp. The first-generation cephalosporins are not effective against the *B. fragilis* group, *Prevotella* spp., and *Porphyromonas* spp. because these organisms all produce cephalosporinase. Regarding the second-generation cephalosporins, there is resistance among the *B. fragilis* strains to cefoxitin, and other *B. fragilis* group members are resistant to cefotetan and cefmetazole. There is variable resistance to third-generation cephalosporins such as ceftizoxime. *B. fragilis* and *B. wadsworthia* are usually resistant.

The carbapenems are a highly effective class of antibiotics for the GNARs, and several have broad-spectrum activity against both aerobic and anaerobic bacteria. Imipenem, meropenem, ertapenem, and doripenem all are US Food and Drug Administration (FDA)–approved carbapenems. Both doripenem and ertapenem provide excellent empirical coverage for complicated intraabdominal infections. However, resistance is emerging, and nonsusceptibility to carbapenems has been reported for *B. fragilis, Fusobacterium* spp., and *Prevotella* spp. isolates.[58] Although chloramphenicol has excellent in vitro activity against GNARs, its attendant toxicities have limited its use.

Clindamycin is a highly effective antibiotic against *Prevotella, Porphyromonas,* and *Fusobacterium* spp. However, there is significant resistance among the *B. fragilis* group (5%–35%) and *Sutterella* spp. (25%–35%).[59] Therefore although clindamycin's activity against aerobic

TABLE 247.2 Antibiotic Sensitivities of Medically Important Gram-Negative Anaerobic Rods

	BACTEROIDES FRAGILIS GROUP	*PREVOTELLA* SPECIES	*PORPHYROMONAS* SPECIES	*FUSOBACTERIUM* SPECIES
Piperacillin-tazobactam	S	S	S	S
Amoxicillin-clavulanic acid	S	S	S	S
Clindamycin	R (V)	S	S	S
Carbapenem	S	S	S	S
Metronidazole	S	S	S	S
Moxifloxacin	S	S	S	S
Penicillin	R	R	R	S

R, Resistant >30%; *S,* sensitive <5%; *V,* variable >5%, <30% sensitivity.

gram-positive cocci makes it an appealing choice for the treatment of polymicrobial infections, its lack of activity against important anaerobes limits its use as a sole drug in anaerobic infections.

Tigecycline, a glycylcycline, is currently the only FDA-approved member of its class. Tigecycline has outstanding activity against a range of clinical isolates including gram-positive and gram-negative anaerobes and gram-positive aerobes.[60] The broad spectrum of tigecycline makes it attractive for empirical therapy for complicated skin, soft tissue, and intraabdominal infections,[61] although the FDA has issued a warning about excessive mortality associated with its use.

Resistance among GNARs to the quinolone antibiotics varies. Although all GNARs are highly susceptible to moxifloxacin, resistance rates for levofloxacin and ciprofloxacin are as high as 50%.[62]

Metronidazole has been a highly effective agent for all GNARs except *Sutterella* spp. for approximately 50 years. Anaerobes convert this prodrug into its active form, which then inhibits their nucleic acid synthesis. The rates of metronidazole resistance among the GNARs remain low—less than 1%. However, there is concern for emerging resistance. The *nim* resistance genes, of which six have been identified, confer resistance to metronidazole by encoding a reductase that prevents the conversion of metronidazole into its active form. A study of more than 1500 *B. fragilis* clinical isolates from Europe detected *nim* gene expression in 2% of samples, raising concern for evolving resistance.[63] Use of metronidazole as a single agent is limited for many infections involving GNARs (e.g., pleuropulmonary infection) because of the resistance of aerobic or microaerophilic streptococci.

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The complete reference list is available online at Expert Consult.

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248 Anaerobic Cocci and Anaerobic Gram-Positive Nonsporulating Bacilli

Eija Könönen

SHORT VIEW SUMMARY

Microbiology and Epidemiology

- Gram-positive anaerobic cocci (*Anaerococcus, Finegoldia, Parvimonas, Peptoniphilus, Peptostreptococcus*) are frequent findings in clinical specimens with anaerobic growth.
- Clinically relevant anaerobic gram-positive nonsporulating bacilli include members of *Cutibacterium* and *Pseudopropionibacterium* (formerly *Propionibacterium*), *Lactobacillus, Actinotignum* (formerly *Actinobaculum*), *Atopobium,* and *Eggerthella*- and *Eubacterium*-like taxa.
- Typical infectious sites with anaerobic cocci or gram-positive anaerobic nonsporulating rods include the central nervous system, eye, oral cavity, respiratory tract, abdomen and intestine, genital tract, bone and joints, wounds, and blood.

Diagnosis

- Proper specimen collection and transport techniques are essential for the successful recovery of strictly anaerobic bacteria.
- Some automated blood culture systems may adversely affect recoveries of gram-positive anaerobic cocci (*Finegoldia magna, Peptostreptococcus anaerobius*) from blood.
- *Actinotignum schaalii* may be overlooked as a uropathogen in routine (aerobic) urine cultures.
- Use of advanced diagnostic methods such as matrix-assisted laser desorption/ionization time-of-flight mass spectrometry, in particular, and sequencing of the 16S ribosomal RNA gene, improves considerably the identification of fastidious organisms and reveals their clinical significance.

Clinical Significance

- Anaerobic bacteremia varies markedly by pathogen; if patients have an underlying condition and are not treated for infection with anaerobes, the mortality rate can be high. In this context, bacteremia with *Lactobacillus* and *Eggerthella*-like species is of special concern.
- Among gram-positive anaerobic cocci, *F. magna* has the highest pathogenic potential. It is involved in skin, soft tissue, and joint infections. *F. magna* and *Peptoniphilus harei* are among the principal species in diabetic foot, venous leg, and decubitus ulcers, which are chronic biofilm-associated infections. *Parvimonas micra* is a common finding in brain abscesses, suggesting an oral source. *Peptostreptococcus anaerobius* is isolated from ulcer and skin specimens of the lower extremities and pus specimens of the genitourinary tract.
- Of gram-positive anaerobic nonsporulating rods, cutaneous propionibacteria, especially *Cutibacterium acnes,* are a clinically relevant finding in cases of implantation of foreign bodies and late postoperative infections. High concentrations of *Atopobium vaginae* are typical for bacterial vaginosis. *Eggerthella lenta* recoveries come from blood, abscesses, wounds, obstetric and genitourinary tract infections, and intraabdominal infections.

Treatment

- Among gram-positive anaerobic cocci, the common resistance to clindamycin, in particular, is a global phenomenon. Some strains resistant to penicillin have been reported. Susceptibilities between different genera and species vary; *P. anaerobius* and *F. magna* are of main concern. *Veillonella* species show increasing resistance to clindamycin.
- For anaerobic gram-positive bacilli, the most active antimicrobial agents are the penicillins and carbapenems. Species- and strain-related resistance to glycopeptides among lactobacilli is common, and the use of vancomycin in the treatment of *Lactobacillus* bacteremia is not advisable. Susceptibility rates of *Eggerthella lenta* to penicillin are consistently poor and to amoxicillin-clavulanate, cefoxitin, and metronidazole good. *Cutibacterium, Bifidobacterium,* and *Lactobacillus* are mostly and *Atopobium, Mobiluncus, Eggerthella,* and *Eubacterium* occasionally resistant to metronidazole. Uropathogenic and bacteremic *Actinotignum* strains show high minimal inhibitory concentration values to trimethoprim-sulfamethoxazole and fluoroquinolones.
- Chronic wounds and foreign body and oral infections are typical biofilm-associated infections for which antimicrobial therapy alone is ineffective, and mechanical intervention is needed in their management.

Both anaerobic cocci and anaerobic gram-positive nonsporulating bacilli belong to the commensal microbiota of the digestive tract, and some are members of the microbiota of the urogenital tract and skin. When the environment changes due to trauma, immunosuppression, or antimicrobial therapy, they can cause damage in a susceptible host and result even in life-threatening infections.[1,2] The major genera of anaerobic cocci and gram-positive anaerobic nonsporulating bacilli are listed in Table 248.1. Other anaerobic bacteria are discussed in Chapters 242 through 247.

TAXONOMY

Gram-positive anaerobic cocci (GPAC) belonging to the family Peptostreptococcaceae in the phylum Firmicutes are strictly anaerobic and vary in their cell size according to the species. This heterogeneous group has confronted remarkable taxonomic revisions in recent years. In 2001, a radical revision among the genus *Peptostreptococcus* was made when saccharolytic species and nonsaccharolytic species, which use peptone as their energy source and produce butyrate as a major metabolite of their glucose fermentation, were separated from each other by reclassifying them as *Anaerococcus* and *Peptoniphilus,* respectively.[3] Since then, several novel species with clinical origin have been added especially to the genus *Peptoniphilus.*[1] In addition, many candidate species of *Anaerococcus* and *Peptoniphilus* are suggested based on a culturomics approach, however, they still wait for validation and further information on their clinical significance. Currently, the original genus *Peptostreptococcus* includes two human species (*P. anaerobius* and *P. stomatis*), while the genera *Finegoldia, Parvimonas,* and *Murdochiella* are each represented by a single species (*F. magna, P. micra,* and *M. asaccharolytica,* respectively) so far.[1] In the genus *Peptococcus, P. niger* has remained the only human species. All of the previously mentioned genera harbor clinically relevant species. Within the family Lachnospiraceae, a strictly anaerobic gram-positive diplococcus, *Ruminococcus gnavus,* has gained interest as being a dominant part of the human gut microbiota but also having clinical relevance.

Among gram-negative anaerobic cocci, the genera *Anaeroglobus, Megasphaera, Negativicoccus,* and *Veillonella* belong to the family

TABLE 248.1 Genera of Anaerobic Bacteria Covered in This Chapter

Gram-Positive Anaerobic Cocci

Anaerococcus
Finegoldia
Murdochiella
Parvimonas
Peptococcus
Peptoniphilus
Peptostreptococcus
Ruminococcus

Gram-Negative Anaerobic Cocci

Acidaminococcus
Anaeroglobus
Megasphaera
Negativicoccus
Veillonella

Gram-Positive Anaerobic Nonsporulating Bacilli
Firmicutes

Bulleidia
Eisenbergiella
Eubacterium
Filifactor
Lactobacillus
Mogibacterium
Moryella
Oribacterium
Pseudoramibacter
Solobacterium
Turicibacter

Actinobacteria

Actinotignum (formerly *Actinobaculum*)
Actinomyces (not covered in this chapter)
Alloscardovia
Atopobium
Bifidobacterium
Catabacter
Collinsella
Cutibacterium (formerly *Propionibacterium*)
Eggerthella
Gordonibacter
Mobiluncus
Pseudopropionibacterium (formerly *Propionibacterium*)

Veillonellaceae, while the genus *Acidaminococcus* was moved to the family Acidaminococcaceae, with both families located in the phylum Firmicutes.[4] Within the genus *Veillonella,* six species (*V. atypica, V. denticariosi, V. dispar, V. parvula, V. rogosae,* and *V. tobetsuensis*) are isolated mainly from the human oral cavity,[5,6] whereas *V. montpellierensis*[7] and the novel *V. seminalis*[8] are found at genital sites. The genus *Acidaminococcus* includes two species (*A. fermentans* and *A. intestini*)[9] and the genera *Anaeroglobus* and *Negativicoccus* one species each (*A. geminatus* and *N. succinivorans,* respectively).[4,10] Of the human *Megasphaera* species, *M. micronuciformis* has been isolated from human clinical specimens.[11] Although the pathogenicity of gram-negative anaerobic cocci is generally considered low, they may be of potential importance especially in mixed infections.

Anaerobic gram-positive nonsporulating bacilli are widely distributed among two phyla, Actinobacteria and Firmicutes.[2] Within the family Actinomycetaceae of the phylum Actinobacteria, the genera *Actinomyces, Actinotignum* (formerly *Actinobaculum*), *Trueperella* (formerly *Arcanobacterium*), and *Varibaculum* include facultatively anaerobic, coccoid or branching rods with a positive Gram stain reaction, while *Mobiluncus* species are motile and strictly anaerobic, curved bacilli with variable Gram reactions.[2,12] Recent attempts to revise the heterogeneous genus *Propionibacterium* led to the transfer of cutaneous propionibacteria (*P. acnes, P. avidum,* and *P. granulosum*) to a novel genus, *Cutibacterium.*[13] Furthermore, *Propionibacterium propionicum* was reclassified as being a single species in the novel genus *Pseudopropionibacterium.* These species are anaerobic and aerotolerant, variable-shaped organisms of which *Cutibacterium acnes,* in particular, can be encountered in clinical material.[2,14] A novel *Propionibacterium, P. namnetense,* which is closely related to *C. acnes,* has clinical relevance.[15] Members of the family Bifidobacteriaceae are strictly anaerobic or occasionally microaerophilic, pleomorphic rods. Human *Bifidobacterium* species reside in the intestine in particular, while two former *Bifidobacterium* species, *Scardovia inopinata* and *Parascardovia denticolens,* and the novel *Scardovia wiggsiae* are mainly isolated from oral sites.[2,16] The closely related *Alloscardovia omnicolens* has been found in various types of human infectious material.[17] The genera *Atopobium* and *Olsenella,* with variable, often coccoid cells, have been created from the so-called anaerobic lactobacilli belonging to the family Coriobacteriaceae.[2] *Atopobium* species with clinical relevance in humans include *A. minutum, A. rimae, A. parvulum,* and *A. vaginae.* In addition, the novel *A. deltae* strain was found as an etiologic agent of sepsis.[18] The genus *Olsenella* includes two human species, *O. uli* and *O. profusa,* isolated from the oral cavity.[2] Although most *Eubacterium* species belong to the Firmicutes, a number of former *Eubacterium* species with a high guanine-cytosine content have been renamed and removed to the phylum Actinobacteria. Among those are *Eggerthella lenta* and *Collinsella aerofaciens* from the gut and *Slackia exigua* from the oral cavity.[2] *Eggerthella sinensis, Paraeggerthella* (formerly *Eggerthella*) *hongkongensis, Gordonibacter pamelaeae,* and the motile and catalase-producing *Catabacter hongkongensis,* in particular, have been found in blood.[19–21]

The other major phylum of gram-positive anaerobic nonsporulating bacilli is the Firmicutes. Within this phylum, the genus *Lactobacillus* constitutes a branch subdivided into several groups, whereas some former *Lactobacillus* species were reclassified and moved to other genera (e.g., *Atopobium* and *Olsenella*).[2] *Eubacterium* and *Eubacterium*-like species are widely distributed among the Firmicutes. Clinically important, mainly in oral infections, are *Eubacterium brachy, E. infirmum, E. minutum, E. nodatum, E. saphenum, E. sulci, Filifactor alocis, Mogibacterium* species, *Pseudoramibacter alactolyticus, Bulleidia extructa,* and *Solobacterium moorei.*[22] Other anaerobic gram-positive nonsporulating bacilli within the Firmicutes with potential clinical relevance are *Turicibacter sanguinis, Oribacterium sinus,* and *Moryella indoligenes,* as well as the novel, gram-variable *Eisenbergiella tayi.*[2,23,24]

Members of the Commensal Microbiota

Among gram-negative anaerobic cocci, *Veillonella* species are considered mainly harmless, or even beneficial, colonizers of the mouth from the early years of life onward.[25] *Acidaminococcus* and *Megasphaera* species reside the human intestine at young age; however, geographic or ethnic differences, or both, in their abundance are significant.[26]

Among GPAC organisms, *R. gnavus* plays an important role as an early colonizer of the human gut.[27] Although members of the genera *Anaerococcus, Finegoldia, Parvimonas, Peptoniphilus,* and *Peptostreptococcus* are commonly present as part of the human microbiota, residing the mouth, intestine, and both the female and male genital tract as well as the surfaces of the eye, nasal cavity, and skin,[1,28–31] only limited knowledge exists about their role as commensal organisms.

Anaerobic gram-positive nonsporulating bacilli belong to the commensal microbiota of the digestive tract, and some members are part of the microbiota of the urogenital tract and skin. The latter environment is widely colonized by cutaneous propionibacteria, *C.* (formerly *P.*) *acnes,* in particular.[14] *Bifidobacterium* and *Collinsella* species are important in the development of the gut microbiota.[26,32] In elderly persons, a decrease in the levels of intestinal *Bifidobacterium* populations is seen with aging.[33] Lactobacilli, such as *Lactobacillus rhamnosus, L. paracasei, L. plantarum,* and *L. salivarius,* form an important component on mucosal surfaces of the gastrointestinal tract.[26,34] The most prevalent lactobacilli in health-associated vaginal biofilms are *L. crispatus, L. gasseri,* and *L. jensenii,* while *L. iners* can also be found in intermediate and dysbiotic states of the vaginal microbiota.[35] Hydrogen peroxide production by these species, except for *L. iners,* may help to control the overgrowth of bacterial vaginosis–associated organisms and to protect from obstetric infections during pregnancy.[36] With variable results, beneficial *Lactobacillus* and *Bifidobacterium* strains have been used as probiotics, aiming to protect the homeostasis or to alter the dysbiotic composition of the microbiota.[37,38]

Clinical Significance

In infectious lesions, the organisms covered in this chapter often appear together with other anaerobic or facultative bacteria in polymicrobial consortia typical at or close to their natural anatomic site. Hematogenous spread of anaerobes from the commensal microbiota and local infections along the local veins and through the general circulation expose the host to systemic infections, such as brain abscesses and endocarditis.[39,40] The clinical significance of anaerobic bacteremia varies markedly by pathogen; if patients have an underlying condition and are not treated with drugs active against anaerobes and drainage when appropriate, the mortality rate can be high.[41–47] According to data from the Mayo Clinic (Rochester, MN), the increased prevalence of anaerobic bacteremia was especially shown for "peptostreptococci", and their increasing resistance to metronidazole was of concern.[41] However, in a population-based monitoring of anaerobic bloodstream infections in the Province of Alberta, Canada, no increase in resistance rates of anaerobes was observed from 2000 to 2008.[45] Moreover, a decrease in the incidence of anaerobic bacteremia was reported from a Belgian university hospital over a 10-year period between the years 2004 and 2013.[46] Besides bacteremia, typical infectious sites where anaerobic cocci or gram-positive anaerobic nonsporulating rods can be involved include the central nervous system, eye, oral cavity, respiratory tract, abdomen and intestine, genital tract, and wounds.[1,2]

GRAM-POSITIVE ANAEROBIC COCCI

With the incorporation of advanced methods into the clinical microbiology arsenal, the impact of GPAC (formerly known as peptostreptococci) as causative agents in a variety of human infections has increased considerably. For example, in anaerobic bacteremias, GPAC organisms are among the most frequent findings,[45,46] and their importance in pathogenic biofilms of chronic ulcers has been emphasized. The most common GPAC species isolated from various types of clinical specimens is *Finegoldia magna,* but *Parvimonas micra, Peptoniphilus harei, Peptostreptococcus anaerobius,* and *Anaerococcus vaginalis* are not rare.[1,39,48]

Anaerococcus

Typical infectious sites for the genus *Anaerococcus* include the female genital tract; *A. hydrogenalis, A. lactolyticus, A. prevotii, A. tetradius,* and *A. vaginalis* have been isolated from vaginal discharges and ovarian abscesses, while *A. octavius* recoveries come from the skin, nasal cavity, and vagina.[3] *A. prevotii, A. vaginalis,* and *A. lactolyticus* are also among the prevalent findings in chronic wound samples.[49–51] In addition, the involvement of *A. prevotii* in mixed surgical infections and skin infections[52,53] and endodontic infections with abscesses[54] has been reported. *A. murdochii* has been recovered from polymicrobial infections, mainly in the lower extremities but also in the neck and a sternal wound.[55,56] Recently, two novel *Anaerococcus* species with clinical relevance have been suggested: *A. degenerii* (four strains isolated from toe tissue, from drainage fluid after rectal carcinoma surgery, from a pelvic abscess, and from an intraabdominal abscess after rectal carcinoma surgery)[57] and *A. nagyae* (three strains isolated from an infectious lesion of the groin, from a nephrourecterectomy wound infection, and from blood).[58]

Finegoldia

Among GPAC organisms, *F. magna* has the highest pathogenic potential and, subsequently, it is found in a variety of human infections.[1,28,48,52] This high pathogenicity is explained by its specific virulence properties.[1,59] Although GPAC species usually appear in polymicrobial infections, *F. magna* can be identified as the only infectious finding.[28,60] Most typically, *F. magna* is involved in wound, soft tissue, and bone and joint infections.[28] In addition, there are reports on its involvement in endocarditis, breast abscesses, pleural empyema, mediastinitis, chronic balanitis, and bacterial vaginosis.[1,61] *F. magna* is one of the principal species in diabetic foot, venous leg, and decubitus ulcers, which are chronic, polymicrobial biofilm-associated skin and skin structure infections.[1,49–51] According to a report of 61 bone and joint infections with anaerobic organisms,[56] *F. magna* was present in 21% of the cases, which all were polymicrobial and were located in the lower extremities, especially in the ankle. There is increasing evidence for *F. magna* as a causative agent in orthopedic implant–associated infections.[56,60,62]

Parvimonas

P. micra is the far most prevalent GPAC species in the mouth, where it is considered a putative pathogen within subgingival biofilms connected to severe periodontitis,[63] infected root canals with or without abscesses,[54,64] failing dental implants,[65] and odontogenic infections.[66] Strikingly, in a Norwegian study, most brain abscesses proved to be infected by *P. micra* or other *Parvimonas* sp.,[39] suggesting an oral origin. Some recoveries come from bacteremias[46] and bone and joint infections.[56,67] Furthermore, *P. micra* has been shown, for the first time, to be capable of causing bacterial meningitis.[68] This organism is also found among the bacterial vaginosis–related microbiota exposing to chronic genital inflammation.[69]

Peptoniphilus

In clinical specimens with GPAC organisms, *P. asaccharolyticus* appears as a frequent finding[28,52]; however, some doubt has been raised about the correct identification of the species.[55,70,71] Instead, a biochemically similar species, *P. harei,* seems to cover a considerable proportion of clinically relevant GPAC findings.[46,48,55,70] *P. asaccharolyticus/P. harei* was frequently detected among 61 bone and joint infections.[56] *P. harei* is one of the primary findings in biofilms of venous leg ulcers, and *P. harei, P. ivorii,* and *P. indolicus* in pressure ulcers.[1,51] *P. lacrimalis* has been isolated from a lacrimal gland abscess[3] and osteoarticular and vaginal specimens[1] but is also detected as a rather common finding in decubitus ulcer samples.[50] Novel *Peptoniphilus* species have been isolated from polymicrobial human infections below the waistline, including *P. coxii* (leg infections, back cyst, flank abscess, endometrial fluid, and tonsil biopsy), *P. duerdenii* (vaginal abscess), *P. gorbachii* (lower extremities), *P. koenoeneniae* (buttock abscess), *P. olsenii* (lower extremities), *P. rhiniditis* (chronic rhinosinusitis), and *P. tyrrelliae* (leg infection, ischial wound, and perirectal abscesses).[72–75]

Peptostreptococcus

P. anaerobius has long been considered an important human pathogen. A similar species, *P. stomatis,* was not described until 2006[76]; therefore the term *P. anaerobius sensu lato* is used when citing earlier literature. There are reports on its involvement in various types of infections all over the body, including the head, respiratory tract, gastrointestinal tract, genitourinary tract, skin and soft tissue, bone and joints, and cardiovascular sites.[28] *P. anaerobius sensu lato* is frequently isolated from abscesses of various anatomic sites, mixed surgical infections, diabetic wounds, and cutaneous wounds caused by dog or cat bites.[28,49,52,77] In the original description of *P. stomatis,* it was considered an oral species, whereas *P. anaerobius* was suggested to harbor sites below the waistline.[76] Both *P. anaerobius* and *P. stomatis* (oral clone CK035) have been detected in infected root canal specimens and bronchoalveolar lavage samples from patients with ventilator-associated pneumonia.[54,78] In a study of 64 *P. anaerobius sensu lato* strains, *P. stomatis* strains originated not only from oropharyngeal but also gastrointestinal specimens, whereas *P. anaerobius* strains originated mainly from ulcer and skin specimens of the lower extremities and pus specimens of the genitourinary tract.[79]

Murdochiella

M. asaccharolytica represents this novel genus as a single species, which has been isolated from an abdominal abscess and a pilonidal cyst aspirate in humans.[80]

Ruminococcus

Some case reports are currently available on *R. gnavus* in human infections.[81–84]

GRAM-NEGATIVE ANAEROBIC COCCI

Acidaminococcus

A. intestini has been isolated from mixed cultures of specimens of the gastrointestinal area in particular, such as abdominal and peritoneal fluids, inguinal and anal abscesses, and the rectum, but also from maxillary necrosis, axillary abscess, and pressure ulcers.[9]

Anaeroglobus

Few reports are available on *A. geminatus.* According to its original description, the characterized strain was recovered from a postoperative

fluid sample collected after gastrectomy.[10] Oral *A. geminatus* recoveries come from persistent root canal infections[85] and subgingival samples of periodontitis patients, especially those suffering from rheumatoid arthritis.[86] Other infectious lesions with *A. geminatus* include pneumonia with empyema and brain abscess.[39,87]

Megasphaera

M. micronuciformis has been isolated from a liver abscess and pus from a fingertip, as part of mixed growth.[4] Other *Megasphaera* recoveries, though less characterized, are from bacterial vaginosis.[69,88]

Negativicoccus

The only species of this genus, *N. succinivorans,* has been isolated from skin and soft tissue specimens in mixed cultures,[4] and from a bacteremia case with an intestinal source.[89]

Veillonella

Although *Veillonella* species are rarely seen as causative infectious agents, they can be involved even in severe infections, such as anaerobic bacteremia and endocarditis, identified as *V. atypica, V. parvula/V. alcalescens, V. dispar,* and *V. montpelliernsis.*[45,46,90] In addition, *V. parvula* has been detected in meningitis and diskitis with secondary bacteremia,[91] osteomyelitis,[92] and prosthetic knee infections,[93] while *V. montpelliernsis* has been isolated from gastric fluid of a newborn and amniotic fluid samples[7]. The novel *V. seminalis* has been recovered from clinical semen samples, Bartholin gland and perianal abscesses, and a groin wound.[8]

GRAM-POSITIVE ANAEROBIC NONSPORULATING RODS

Propionibacteria

Cutibacterium (*Propionibacterium*) *acnes* is currently recognized as an important pathogen in bone and joint infections, especially in joints with implants.[14,56,94] In a study dealing with 276 patients, the shoulder had a propensity for developing *C. acnes* arthritis compared to lower limbs.[94] This may make sense, because the chest area has a high content of sebaceous glands where cutaneous propionibacteria dominate. Postoperative spondylodiskitis (spinal osteomyelitis) with an involvement of *C. acnes* differs from that caused by aerobic bacteria; it mainly appears in middle-aged subjects, it is rarely complicated by epidural abscess, and there is usually a long incubation period between the preceding invasive operation and infection.[95] Moreover, in the case of anaerobic spondylodiskitis, blood cultures often remained negative. *C. acnes* has been detected in connection to intervertebral disk degeneration,[14,96] and it was possible to demonstrate the high prevalence of *C. acnes* biofilms in the disk tissue.[96] Of the cutaneous propionibacteria, *Cutibacterium* (*Propionibacterium*) *avidum* has also caused attention as a potential pathogen; recent reports have connected this organism to prosthetic hip joint infections[97,98] and soft tissue infections[99–101] after surgical procedures at close proximity to a deep skin crease. Infectious cases with *Cutibacterium* (*Propionibacterium*) *granulosum* have been reported in infective endocarditis in a native mitral valve[102] and in a prosthetic hip.[103] The novel *Propionibacterium namnetense,* which is closely related to *C. acnes,* has been isolated from infected bone (tibia) and surrounding tissue.[15] A few cases of severe skull base osteomyelitis with *C. acnes* as a single isolate have been reported recently.[104] Implantation of medical devices such as intraocular lenses (see Chapter 115), spinal and ventriculoperitoneal shunts (see Chapter 92), and prosthetic heart valves exposes the recipient to infections by cutaneous propionibacteria.[14] Indeed, most endocarditis cases caused by *C. acnes* involve prosthetic cardiac devices in middle-aged men.[105] There is also a risk for further destruction, such as aortic root abscess.[106] Typically, *C. acnes* is isolated from sebaceous follicles, causing lesions of acne vulgaris, where the susceptibility of the host seems to be a dominant factor.[107] Specimens from central nervous system infections, endodontic infections, and infected dog and cat bite wounds can harbor propionibacteria, usually *C. acnes* or, in few cases, *C. granulosum.*[64,77,108] In addition, *Propionibacterium acidifaciens* has been detected in cariotic teeth and persistent endodontic infections.[64,109] *Pseudopropionibacterium* (*Propionibacterium*) *propionicum* resembles phenotypically *Actinomyces israelii,* and is involved in actinomycotic lesions and abscesses, infections of the eye, and endodontic infections.[12]

Lactobacilli

Lactobacillus species not only are beneficial organisms but can act as causative agents in serious infections, especially in immunocompromised individuals with prolonged hospitalization.[40,44,45,110–112] Among the clinically most relevant species are *L. rhamnosus, L. salivarius, L. paracasei, L. casei, L. fermentum,* and *L. acidophilus.* Intraabdominal infections form the main focus of lactobacilli.[44] Their presence in blood specimens is of medical importance because bacteremia and endocarditis with involvement of lactobacilli carry a relatively high mortality rate.[40,44,45,111] *L. rhamnosus,* which is typically vancomycin resistant, is among the most frequent *Lactobacillus* findings in blood.[40,110,111,113] A report from Taiwan, however, found *L. salivarius* as the dominant species in bacteremias caused by lactobacilli.[44] Whether probiotic *Lactobacillus* strains could be of harm has been speculated.[112,114] In a recent study,[115] it was shown that the probiotic strain *L. rhamnosus* GG differs from clinical blood isolates, of which some were able to form biofilms or to induce platelet aggregation. Lactobacilli are among major recoveries from advanced dental caries lesions,[113] and, interestingly, a dental procedure or condition was suggested as a potential predisposing factor in half of the cases of *Lactobacillus* endocarditis.[40] In addition to bacteremia and endocarditis, lactobacilli can be found in infections such as meningitis, empyema, cholecystitis, peritonitis, pyelonephritis, and prosthetic knee infection.[113] Different from other vaginal *Lactobacillus* species, *L. iners* has been observed to dominate in dysbiotic vaginal communities that are susceptible to bacterial vaginosis.[35]

Bifidobacteria

Except for the involvement of *Bifidobacterium* sp. in dental caries, they are generally considered nonpathogenic, and some strains (e.g., *B. breve, B. longum*) are used as probiotics. In the current literature, however, there is an increasing awareness of the potential pathogenicity of bifidobacteria. In pediatric and adult bacteremic cases, with or without preceding probiotic use, the recognized species were *B. infantis, B. adolescentis, B. breve, B. longum,* and *B. dentium* (in the older literature named as *B. eriksonii*).[116] Several types of infections caused by bifidobacteria have been reported.[117] Occasional infectious findings include also *B. scardovii.*[118] In the closely related genus *Alloscardovia, A. omnicolens* was named referring to its presence everywhere in the human body.[17] Clinical *A. omnicolens* isolates originate from blood, urine, genital, pulmonary, oral, and abscess specimens.[17,119] In a study in which 15 putative bifidobacterial isolates from sterile sites or those present in significant numbers were identified to the species level, four species were identified: *A. omnicolens, B. breve, B. longum,* and *B. scardovii* from the gastrointestinal or genitourinary tract, and *B. breve* from blood.[120] *Scardovia wiggsiae,* a novel species, has been linked to severe early childhood caries.[121]

Atopobium and *Olsenella*

A. vaginae is involved in infections of the female genital tract. High concentrations of *A. vaginae* are typical for bacterial vaginosis and, thus, a diagnostically valuable marker of this dysbiotic state.[88] Moreover, *A. vaginae* has been reported from uterine endometritis and bacteremia cases connected to invasive infections of the female genital tract.[122] *A. minutum* has been isolated from various infections of the lower part of the body, *A. parvulum* from respiratory specimens and endodontic infections, and *A. rimae* from severe odontogenic infections.[22,123] *Olsenella* species, especially *O. uli,* have been detected in various oral infections.[54,123] In addition, *A. rimae* and *O. uli* have been reported as causative organisms in clinically significant bacteremia.[110,124]

Actinomyces-Like Organisms

The genus *Actinomyces* is discussed in the chapter on actinomycosis (see Chapter 254). Of the three species of the genus *Actinotignum* (formerly *Actinobaculum*), *A. schaalii* has emerged as an important uropathogen in the elderly, but pediatric cases have also been reported.[12] Cystitis, pyelonephritis, and urosepsis are typical infections caused by

A. schaalii. Most cases of urosepsis have been patients with a predisposing condition, such as renal stones and lithotripsy or other instrumentation. In addition, *A. schaalii* has been found in blood and abscess specimens as well as those from cellulitis, spondylodiskitis, bladder necrosis, epididymitis, and endocarditis.[12,125–127] *Actinotignum* bacteremia patients are typically older males, and a relatively high mortality (16%) has been reported.[127] All strains of the two *Varibaculum* species, *V. cambriense* and the novel *V. anthropi,* have been isolated from clinical specimens.[12,128] Typically, they were involved in abscesses at various body sites, but also in infections of the female genital tract.

Mobiluncus

Of the two *Mobiluncus* species, *M. curtisii* appears to be more virulent than *M. mulieris. M. curtisii* has been connected to bacterial vaginosis, in which its presence is seen as vibrio-like organisms in smears of vaginal fluid and the treatment failure as persistence of the organism.[88] In addition, *M. curtisii* has been occasionally isolated from endometrial smears and pus specimens of the female genital tract.[129] Some reports on extragenital infections and bacteremias with *Mobiluncus* species have been presented.[130]

Eggerthella and Related Species

This group of closely related species is known to possess particular pathogenic potential, causing severe invasive infections. Of these, *Eggerthella lenta* is most common and has been isolated especially from mono- and polymicrobial bacteremias, often with a significant mortality rate.[43,131,132] Other clinically relevant isolations come from abscesses, wounds, spondylodiskitis, osteomyelitis, obstetric and genitourinary tract infections, and intraabdominal infections.[133–135] A retrospective analysis of 33 *E. lenta* bacteremias (January 2000–September 2013) was performed in a large health care network in Australia.[132] A variety of comorbidities at intraabdominal sites, in particular, were found as a background factor, underlining the importance of abdominal diagnostics when *E. lenta* is identified in blood. This was supported by an analysis from a Swedish tertiary hospital with 18 *E. lenta* bloodstream infections.[136] It was recently shown that *E. lenta* can be found in blood of oncologic patients with advanced disease but without infection, indicating translocation of this gut bacterium due to damage of the intestinal barrier.[137] In a tertiary medical center in Taiwan, all "*Eubacterium*"-associated bacteremias in the years 2001–2010 were analyzed[43]; of the 16 patients found, 7 harbored *E. lenta* and 3 *Paraeggerthella hongkongensis;* furthermore, 9 had polymicrobial bacteremia in which *Bacteroides* species were the most common concomitant findings. The gastrointestinal and female genital tracts were suggested as the main entries of these bacteria to blood.[43] In a hospital in Detroit, 25 bacteremia cases with *E. lenta* from 1999 to 2010 were retrospectively reviewed; half of the bacteremias were monobacterial, and the main sources were abdominal and complicated soft tissue infections.[131] Also, *Catabacter hongkongensis* and *Gordonibacter pamelaeae* have been found in blood samples in pure culture, and bacteremia had mainly a gastrointestinal source.[42,138,139] An oral species, *Slackia exigua,* is found in odontogenic and human bite infections,[22,140] but can seemingly be translocated to the pulmonary area; it was reported to be the causative organism of a severe empyema in a patient with multiple cariotic lesions in her teeth.[141] Other extraoral infections with *S. exigua* include bacteremia, and abscesses and wounds at various sites of the body.[46,142]

Eubacterium and Related Taxa

Various *Eubacterium* sp. (e.g., *E. brachy, E. nodatum, E. saburreum,* and *E. sulci*) and related species (e.g., *Filifactor alocis, Mogibacterium timidum, Mogibacterium vescum,* and *Pseudoramibacter alactolyticus*) residing the oral cavity have been recovered from periodontal, endodontic, and odontogenic infectious lesions.[2,22,54] Conceivably, organisms of this group are among the anaerobic recoveries from infected human bite wounds.[140] In a Norwegian multicenter study, *E. brachy* was among the main findings in 37 spontaneous brain abscesses, and *F. alocis* also was found in two cases.[39] A recent report presented a case initially suspected as thoracic actinomycosis due to its chronicity but where *E. brachy* proved to be the causative organism.[143] In addition to being isolated from infections in the upper part of the body, *E. nodatum* has been found in infections of the female genital tract.[144] *E. limosum, E. callanderi,* and *E. tenue* have been detected in clinically significant bacteremias.[43,110] Also, few bacteremia cases of *Solobacterium moorei* have been reported in connection to various comorbidities.[145] Some isolates of *S. moorei* come from wound infections together with other bacteria, especially anaerobes.[146] *Bulleidia extructa* has been found in odontogenic infections, a brain abscess, and a periprosthetic hip joint infection.[22,39,147] Among other clinically relevant Firmicutes taxa, *Turicibacter sanguinis* has been isolated from human blood, *Oribacterium sinus* from pus of a human sinus, *Moryella indoligenes* from abscesses below the waistline, and *Eisenbergiella tayi* from blood and appendix tissue.[2,23,24,148]

MICROBIOLOGIC ASPECTS

Proper specimen collection and transport techniques are essential for the successful recovery of strictly anaerobic bacteria. Avoidance of contamination by commensals of the surrounding skin, mucous membranes, and nonsterile secretions is crucial; therefore mucosal or cutaneous swabs are not appropriate for specimen collection, although they are still used.[149] Instead, samples collected by tissue biopsy, wound curettage, or aspiration should be used as specimens. Transport media should be specifically designed for the survival of anaerobic bacteria.

It is likely that the involvement of anaerobic bacteria in infections is underestimated because many of these organisms are slow-growing and fastidious. Noteworthy is that the automated blood culture system may affect some GPAC recoveries from blood.[1] For instance, *Peptostreptococcus* species are sensitive to sodium polyanethol sulfonate,[79] which is a common component in automated blood culture sets. Also, the detection of *Finegoldia magna* may be affected by some commonly used blood culture systems.[1] Another concern is the potential misinterpretation of isolated organisms such as cutaneous propionibacteria from blood specimens or *Actinotignum* from urine as insignificant contaminants.[105,126]

In the laboratory, clinical specimens should be processed without delay using rich, nonselective media and prolonged incubation (up to 10–14 days).[2] In implant-associated infections, sonication of the removed implant (or part of it) and multiple conventional cultures are recommended.[14] In mixed cultures, many anaerobic organisms can be easily overgrown by facultatives, leading to biased estimates of their true numbers and clinical significance. Currently, *Actinotignum schaalii* is considered a significant uropathogen[12] but it is often missed in routine (aerobic) urine cultures; therefore advanced methods are required for blood isolates from urosepsis. Notably, for some taxa within the phylum Actinobacteria, polymerase chain reaction–based methods with standard primers may result in less successful detection than anaerobic culture. Accurate identification of anaerobic cocci and *Eubacterium*-like organisms, in particular, is limited if only phenotypic tests are used.[22,55] Diagnostic methods such as matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS), in particular, and sequencing of the 16S ribosomal RNA gene are increasingly used for identification in clinical microbiology laboratories. With these methods, clinically relevant but fastidious organisms (e.g., *Eggerthella lenta* or *Actinotignum schaalii*) can be reliably identified.[126,132,136] However, refined technical procedures as well as comprehensive databases are needed for an excellent performance of MALDI-TOF MS for gram-positive anaerobes; for instance, *Bifidobacterium longum, Finegoldia magna,* and *Peptoniphilus ivorii* have been shown to require specific extraction steps in MALDI-TOF MS,[150] while an expanded number of main spectral profiles has improved the correct identification of GPAC organisms.[151] Adoption of culture-independent methods, together with an increased awareness regarding the infectious role of anaerobes, on one hand, will help to reveal the clinical relevance of fastidious organisms covered in this chapter. On the other hand, the more precise identification brings a challenge to clinicians to recognize a variety of bacterial names given for findings in clinical specimens.

TREATMENT

Initial diagnosis of infection and choice of antimicrobial therapy is often based on empirical information, while awaiting laboratory test results. Susceptibility profiles can significantly differ between countries or even between institutions; therefore it would be of benefit to conduct periodically local susceptibility surveys for clinically relevant anaerobes

to guide the therapy. Unfortunately, hospital laboratories seldom perform susceptibility testing for anaerobic isolates and, if performed, very few antimicrobials are tested.[149]

Tables 248.2 and 248.3 present susceptibilities of major gram-positive anaerobic cocci and bacilli to a number of antimicrobials. Among GPAC organisms, the common resistance to clindamycin, in particular, is a global phenomenon.[152–157] In addition, some GPAC strains resistant to penicillin have been reported.[48,153,154] Susceptibilities between different genera or even closely related species vary; for instance, *Peptostreptococcus anaerobius* constantly shows higher minimal inhibitory concentration (MIC) values when compared to those of *Peptostreptococcus stomatis*.[79] Also the susceptibility of *Finegoldia magna* as the most common GPAC recovery from infectious material is of special concern.[1,59,153] Because metronidazole is the drug of choice in many anaerobic infections, metronidazole-resistant strains can impair the treatment outcome. In a multicenter survey from the United States, 98% and 96% of GPAC organisms were susceptible to metronidazole according to the Clinical and Laboratory Standards Institute breakpoints (MIC ≤8 μg/mL), in the years 2007–2009 and 2010–2012, respectively.[158] In contrast, nearly 30% of the 14 strains identified as GPAC were interpreted as resistant to metronidazole (MIC >4 μg/mL) using the European Committee on Antimicrobial Susceptibility Testing (EUCAST) breakpoint in a Croatian report.[159] A high prevalence of nitroimidazole resistance genes has been found among GPAC species, especially within *P. anaerobius* isolates, and two highly resistant strains of *F. magna* were also detected.[160] Quinolone resistance of *F. magna* is common globally, and two *F. magna* strains resistant to chloramphenicol have been reported in Korea.[59] Among gram-negative cocci, high resistance rates to penicillin have been reported for *Veillonella* species.[152,156,158] Between the periods of 2007–2009 and 2010–2012 reported from the United States, clindamycin resistance increased from 7% to 34%, respectively, among *Veillonella*.[158]

In general, the antimicrobial agents most active against anaerobic gram-positive bacilli are the penicillins and carbapenems. It is notable that species- and strain-related resistance to glycopeptides is frequent among the genus *Lactobacillus*.[40,44,113] Therefore the use of vancomycin for the treatment of bacteremia or infections with involvement of lactobacilli is not advisable. Also cephalosporins are not consistently effective in the treatment of *Lactobacillus* bacteremia. Bifidobacteria and the closely related *Alloscardovia omnicolens,* a rare pathogen from various clinical specimens, are susceptible to β-lactams.[119,152,161] According to data on uropathogenic and bacteremic *Actinotignum* strains, MIC values to β-lactams and vancomycin are low but those to trimethoprim-sulfamethoxazole and fluoroquinolones, especially ciprofloxacin, are high.[125,127] Susceptibility rates of *Eggerthella lenta* to penicillin were consistently poor and those to amoxicillin-clavulanate, cefoxitin, and metronidazole good, while other resistance rates varied considerably between studies—for example, those for piperacillin-tazobactam ranged from 0 to 83%, and for clindamycin from 0 to 18%.[132,136,152] Although clindamycin has good bone penetration, its use in bone- and joint-related infections caused by *Cutibacterium acnes* may be of concern due to its increasing resistance.[14] Nearly complete resistance of *C. acnes* to metronidazole has been shown to be independent of geography,[14,152,156–158] and metronidazole-resistant strains are common also among other facultatively anaerobic/microaerophilic organisms such as *Bifidobacterium* and *Lactobacillus*.[113,152,161]

Diverse bacteriology of intraabdominal infections recalls antimicrobial therapy that covers both aerobic and anaerobic bacteria, such as cephalosporin or fluoroquinolone combined with metronidazole or monotherapy with carbapenem or tigecycline.[162] Some novel drugs, among those tigecycline and linezolid, exhibit potential activities against anaerobic bacteria; however, concern has been raised regarding the efficacy and safety issues of tigecycline therapy.[163] When anaerobic bacteria are involved in biofilm infections, as is the case typically with oral infections, various chronic wounds, and foreign bodies, antimicrobial therapy alone is ineffective despite in vitro susceptibility of the potential causative organism(s) to antibiotics.[164] Due to biofilm structure, mechanical intervention (drainage, surgery) or removal of foreign bodies is needed in the management of these infections.

TABLE 248.2 Susceptibilities of Anaerobic Cocci to Antimicrobials

	ANAEROCOCCUS	*FINEGOLDIA*	*PEPTONIPHILUS*	*PEPTOSTREPTOCOCCUS*	*PARVIMONAS*	*VEILLONELLA*
Penicillin	≤0.002–1	0.016–1	0.016–4	0.016–16	0.016–0.25	≤0.002–1
Amoxicillin	ND	<0.016–0.5	<0.016–0.125	ND	<0.016–2	0.023–2
Amoxicillin-clavulanate	0.016–0.5	0.016–0.5	0.016–0.25	0.016–8	0.016–0.125	<0.016–16
Piperacillin-tazobactam	≤0.015–0.125	0.015–0.25	ND	ND	≤0.015–0.015	≤0.015–1
Cefoxitin	≤0.015–0.5	0.06–1	ND	ND	≤0.015–2	0.12–8
Imipenem	0.002–0.125	0.002–0.25	0.002–0.125	0.002–1	0.002–0.125	0.004–2
Meropenem	ND	0.03–0.12	ND	ND	0.004–0.12	0.015–2
Clindamycin	≤0.015–>256	0.016–>256	0.016–>256	0.016–256	≤0.015–>256	≤0.015–256
Metronidazole	0.016–0.5	0.016–4	0.016–2	0.016–1	≤0.015–1	<0.016–4

ND, No data available.
[a]Range of minimal inhibitory concentrations (MICs;mg/L) of tested strains to indicated antimicrobial agents.[152,155,156]

TABLE 248.3 Susceptibilities of Gram-Positive Anaerobic Bacilli to Antimicrobials

	ACTINOTIGNUM	*ALLOSCARDOVIA*	*BIFIDOBACTERIUM*	*COLLINSELLA*	*EGGERTHELLA*	*EUBACTERIUM*	*LACTOBACILLUS* (VANCOMYCIN-RESISTANT)	*LACTOBACILLUS* (VANCOMYCIN-SUSCEPTIBLE)	*PSEUDOPROPIONIBACTERIUM*
Penicillin	0.004–0.5	≤0.01–0.12	0.015–2	0.002–8	0.004–4	0.003–2	0.125–>32	≤0.06–0.5	≤0.004–2
Amoxicillin-clavulanate	ND	0.06–0.5	ND	ND	≤0.06–2	ND	0.12–>32	0.25–0.5	ND
Piperacillin-tazobactam	ND	0.12–0.5	≤0.015–0.5	0.015–0.125	≤0.03–64	≤0.015–0.03	0.12–>64	0.25–4	0.015–2
Cefoxitin	≤0.015–1	ND	0.12–>256	ND	0.15–16	0.06–>256	16–>128	8–>128	0.015–4
Ertapenem	ND	ND	ND	ND	0.06–2	ND	0.5–>16	0.5–8	≤0.06–0.75
Imipenem	ND	≤0.01–0.12	ND	ND	ND	ND	≤0.015–16	0.03–1	≤0.03–2
Meropenem	0.004–0.06	ND	0.015–2	≤0.002–0.12	≤0.06–4	0.007–0.12	ND	ND	0.06–1.5
Vancomycin	ND	0.12–1	ND	ND	0.25–8	ND	4–>128	0.5–2	0.016–2
Daptomycin	ND	≤0.01–16	ND	ND	ND	ND	0.125–4	0.5–>32	0.125–2
Linezolid	ND	0.12–1	ND	ND	ND	ND	1–8	1–>32	0.016–2
Tigecycline	ND	≤0.01–0.06	ND	ND	≤0.06–1	ND	ND	ND	0.016–0.064
Moxifloxacin	ND	0.06–4	ND	ND	0.03–>32	ND	ND	ND	0.125–0.5
Levofloxacin	ND	0.25–>32	ND	ND	ND	ND	0.25–>32	0.5–>32	0.125–≥256
Clindamycin	≤0.015–>256	≤0.01–>256	≤0.015–8	<0.015–64	≤0.015–>256	≤0.012–32	≤0.03–0.5	≤0.03–4	≤0.016–>256
Metronidazole	0.06–>256	≥32	0.12–>256	0.03–>256	≤0.016–>64	≤0.015–0.5	ND	ND	0.06–>256

ND, No data available.
[a]Range of minimal inhibitory concentrations (MICs;mg/L) of tested strains to indicated antimicrobial agents. [14,44,113,119,132,152]