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244 Tetanus (*Clostridium tetani*)

Torrey Boland Birch and Thomas P. Bleck

SHORT VIEW SUMMARY

History
- Tetanus is a nervous system disease caused by the toxin tetanospasmin produced by *Clostridium tetani*.
- Vaccination for tetanus has been widely available in developed countries since the 1940s.

Epidemiology
- Acute injury and injection drug use are risk factors for tetanus.
- Tetanus is rare in developed countries due to the availability of effective vaccines.
- In developing countries neonatal tetanus can occur when there is failure of aseptic technique and mothers are inadequately immunized.
- In the United States tetanus is more common in people older than 65 years, likely due to waning immunity.

Pathogenesis
- *C. tetani* is an obligately anaerobic bacillus that produces two toxins: tetanospasmin and tetanolysin.
- It develops a terminal spore that is extremely stable in the environment, retaining the ability to germinate and cause disease indefinitely.
- Tetanospasmin enters the nervous system through the lower motor neurons and is carried to the brain and spinal cord via retrograde transport.
- Tetanospasmin prevents release of neurotransmitters from inhibitory cells leading to increased muscle tone and a hypersympathetic state.

Clinical Manifestations
- Tetanus is divided into four clinical types: *generalized, localized, cephalic,* and *neonatal*.
- Hallmarks of generalized tetanus include trismus (lockjaw, or masseter rigidity) and risus sardonicus (increased tone in the orbicularis oris)

Diagnosis
- Diagnosis of tetanus relies on history and examination findings.
- Culturing *C. tetani* is difficult and not helpful.

Treatment
- The airway must be secured at the time of presentation.
- Benzodiazepines provide the mainstay of symptomatic therapy.
- Passive immunization with human tetanus immune globulin (HTIG) shortens the disease course and may lessen disease severity.
- Antibiotic therapy with metronidazole may improve outcomes.

Prevention
- All children should be vaccinated against tetanus through the diphtheria-tetanus-acellular pertussis (DTaP) vaccine series.
- All pregnant women should receive a dose of the tetanus-reduced diphtheria toxoid-acellular pertussis (Tdap) vaccine with each pregnancy
- Adults should receive a tetanus booster (Td) every 10 years. One of the Td doses should be replaced with Tdap.
- Wound management of minor, clean wounds should include completion of tetanus immunization if incomplete or a booster dose of vaccine (Td) if the last dose was given more than 10 years before. Patients with serious contaminated wounds should also receive HTIG.

HISTORY

Tetanus was well known to the ancients; descriptions by Egyptian and Greek physicians survive to the present. They recognized the frequent relationship between injuries and the subsequent development of fatal spasms. Gowers[1] provided the quintessential description of tetanus in 1888.

Tetanus is a disease of the nervous system characterized by persistent tonic spasm, with violent brief exacerbations. The spasm almost always commences in the muscles of the neck and jaw, causing closure of the jaws (trismus, lockjaw), and involves the muscles of the trunk more than those of the limbs. It is always acute in onset, and a very large proportion of those affected die.[1]

Nicolaier[2] isolated a strychnine-like toxin from anaerobic soil bacteria in 1884. Six years later, Behring and Kitasato[3] described active immunization with tetanus toxoid. Inactivation of tetanus toxin was developed in the early 20th century and was widely used for treatment and prevention by the 1940s. This latter discovery should have reduced tetanus to a historical curiosity, but we still fail to fulfill this promise.

EPIDEMIOLOGY

Between 2001 and 2008 the Centers for Disease Control and Prevention (CDC) received reports of 233 tetanus cases, with an overall annual incidence of 0.10 cases per 1 million persons and an incidence of 0.23 cases per 1 million persons 65 years or older.[4] Data through 2008 are summarized in Fig. 244.1. Globally, 13,505 cases of tetanus were reported in 2016.[5] United States and global statistics likely represent underreporting. Most reported cases are in patients older than 60 years,[6] indicating that waning immunity is an important risk factor.[7] This may be a particularly serious problem in older women.[8,9] Changes in patterns of immigration may increase the number of unimmunized or inadequately immunized patients presenting for care in developed countries.[10] Injection drug abuse places patients at risk for tetanus,[11] as do other potentially unsterile practices that allow inoculation of spores.[12]

Causes of Tetanus

C. tetani germinates in low oxygen conditions, particularly in wounds. Acute injuries account for about 70% of US cases, evenly divided between punctures and lacerations.[13] Other identifiable conditions are noted in 23%, leaving about 7% of cases without an apparent source. Other studies cite rates of cryptogenic tetanus as high as 23%. Outbreaks of tetanus are common after natural disasters in developing countries, when people are likely to suffer from open wounds, fractures, and crush injuries.[14] Neonatal tetanus is rare in the United States.

Mortality

In developing countries, mortality rates due to tetanus are as high as 28 per 100,000. Until recently, primary tetanus immunization programs in these countries were ineffective. As a result, 800,000 to 1 million annual deaths were attributed to tetanus during the 1980s.[15] Two-thirds of cases worldwide occurred in sub-Saharan Africa, where more than

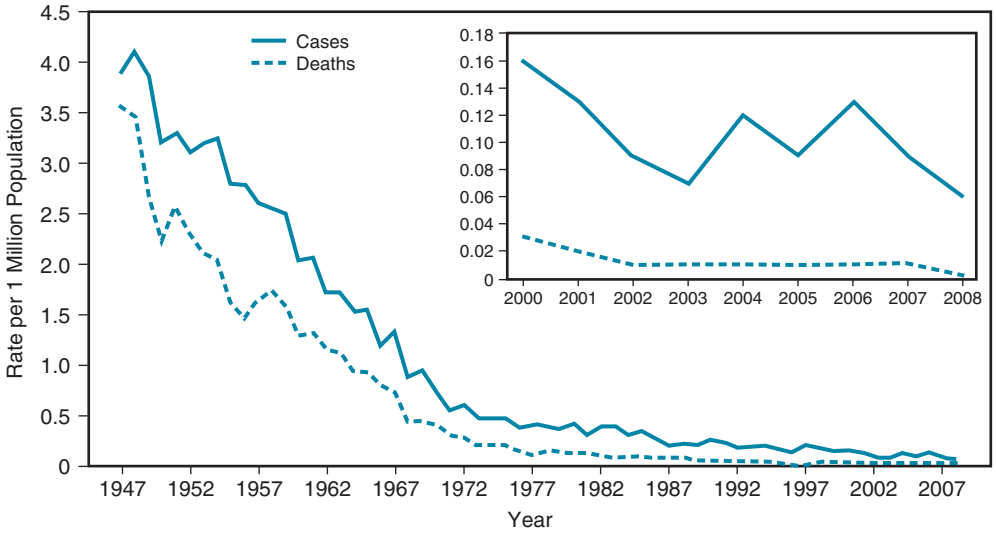

FIG. 244.1 Reported cases and deaths from tetanus in the United States, 1947–2008. *(Data from Centers for Disease Control and Prevention. Tetanus surveillance—United States, 2001–2008.* MMWR Morb Mortal Wkly Rep. *2011;60:365–369.)*

40% of tetanus is a result of neonatal infection[15,16]; nearly one-third of these infants were born to mothers of a previously afflicted child, highlighting a failure to immunize.[17]

In 1989 a worldwide commitment to the elimination of neonatal tetanus by the World Health Assembly[18,19] resulted in a decline of more than 50%, and a resurgent effort in 1999,[20] the Maternal and Neonatal Tetanus Elimination Program,[21] met with additional success. In 2015 the World Health Organization (WHO) estimated that there were 34,019 neonatal deaths due to tetanus, representing a 96% reduction in neonatal tetanus deaths compared with the 1980s.[22] The Global Immunization Vision and Strategy, launched by WHO and United Nations Children's Fund in 2005, continues to target tetanus as a preventable cause of neonatal death by promoting routine tetanus toxoid administration in hard-to-reach, previously underserved areas.[23] As of 2014, it was estimated that globally, 86% of infants had received at least three doses of the diphtheria-tetanus-pertussis vaccine.[24]

MICROBIOLOGY OF *CLOSTRIDIUM TETANI*

Clostridium tetani is an obligately anaerobic bacillus that is gram positive in fresh cultures but may have variable staining in older cultures or tissue samples.[25] It is present in soil, especially heavily manured soil, and in the intestinal tract and feces of various animals. It is believed to be transiently present in the human gastrointestinal (GI) tract, perhaps dependent on recent ingestion.[26] The complete genome of the organism has been sequenced, and its products were recently compared with other clostridia.[27] During growth the bacilli possess abundant flagella and are sluggishly motile. Two toxins, tetanospasmin (commonly called *tetanus toxin*) and tetanolysin, are produced during this phase. Tetanospasmin is encoded on a plasmid that is present in all toxigenic strains.[28] Tetanolysin is of uncertain importance in the pathogenesis of tetanus. Mature organisms lose their flagella, develop a terminal spore, and begin to resemble a squash racquet (Fig. 244.2).[29] The spores are extremely stable in the environment, retaining the ability to germinate and cause disease indefinitely. They withstand exposure to ethanol, phenol, or formalin but can be rendered noninfectious by iodine, glutaraldehyde, hydrogen peroxide, or autoclaving at 121°C and 103 kilopascals (15 psi) for 15 minutes. Growth in culture is optimal at 37°C under strictly anaerobic conditions, but culture results are of no diagnostic value. Antibiotic sensitivity is discussed later.

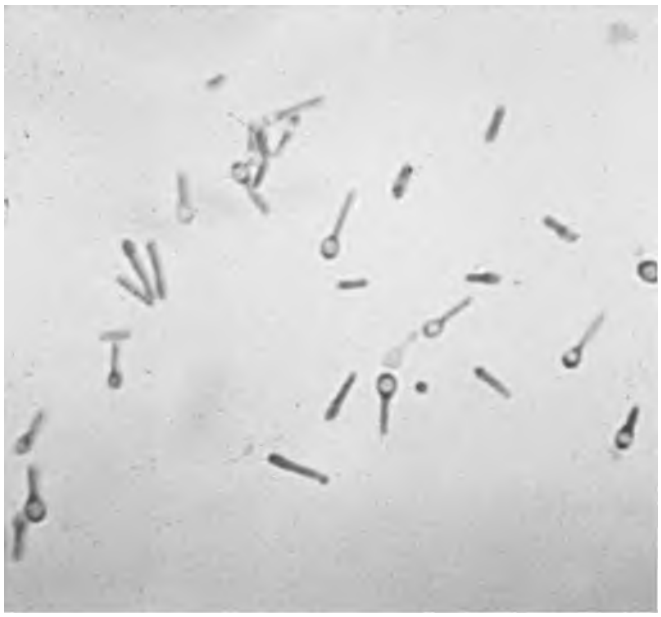

FIG. 244.2 Gram stain of a culture of *Clostridium tetani*. *(From Bleck TP, Brauner JS. Tetanus. In: Scheld WM, Whitley RJ, Marra CM, eds.* Infections of the Central Nervous System. *3rd ed. New York: Lippincott Williams & Wilkins; 2004:625–648. Courtesy Paul C. Schrechenberger, PhD, and Alex Kuritza, PhD.)*

PATHOGENESIS

The clostridial toxins that produce both tetanus and botulism are similar in structure and function despite the almost diametrically opposed clinical manifestations of the diseases. These toxins are zinc-dependent matrix metalloproteinases, a category encompassing a diverse group of enzymes ranging from normal human cellular constituents necessary for cellular remodeling,[30] through determinants of neoplastic cell function,[31] to exotoxins of other microorganisms such as *Bacteroides fragilis*.[32] Tetanospasmin is synthesized as a single 151-kilodaltons (kDa) chain that is cleaved extracellularly by a bacterial protease into a 100-kDa heavy chain and a 50-kDa light chain (fragment A), which remain connected by a disulfide bridge.[33] The heavy chain can be further divided into fragments B and C by pepsin. The heavy chain appears to mediate binding to cell surface receptors and transport proteins, whereas the light chain produces the presynaptic inhibition of transmitter release that produces clinical tetanus. The nature of the receptor to which tetanospasmin binds, previously thought to be a ganglioside, has been debated.[34] Recent research suggests that the extracellular matrix proteins nidogen-1 and nidogen-2 serve as the tetanospasmin receptor.[35] The

toxin enters the nervous system primarily through the presynaptic terminals of lower motor neurons, where it can produce local failure of neuromuscular transmission. It then exploits the retrograde axonal transport system and is carried to the cell bodies of these neurons in the brainstem and spinal cord, where it expresses its major pathogenic action.[36]

Once the toxin enters the central nervous system (CNS), it diffuses to the terminals of inhibitory cells, including both local glycinergic interneurons and descending γ-aminobutyric acid–ergic (GABAergic) neurons from the brainstem. The toxin degrades synaptobrevin, a protein required for docking of neurotransmitter vesicles, with their release site on the presynaptic membrane.[37] By preventing transmitter release from these cells, tetanospasmin leaves the motor neurons without inhibition. This produces muscular rigidity by raising the resting firing rate of motor neurons and also generates spasms by failing to limit reflex responses to afferent stimuli. Excitatory transmitter release in the spinal cord can also be impaired, but the toxin appears to have greater affinity for the inhibitory systems. The autonomic nervous system is affected as well; this is predominantly manifested as a hypersympathetic state induced by failure to inhibit adrenal release of catecholamines.

Toxin binding appears to be an irreversible event. At the neuromuscular junction, initial recovery from botulism depends on sprouting a new axon terminal; this is probably the case at other affected synapses as well. Later, the new synapses are removed when the original ones reestablish their connections.[38]

CLINICAL MANIFESTATIONS

Tetanus is classically divided into four clinical types: *generalized, localized, cephalic,* and *neonatal.* These are valuable diagnostic and prognostic distinctions but reflect host factors and the site of inoculation rather than differences in toxin action. Terms describing the initial stages of tetanus include the *incubation period* (time from inoculation to the first symptom) and the *period of onset* (time from the first symptom to the first generalized spasm). The shorter these periods, the worse the prognosis.[39] The incubation period ranges from 3 to 21 days.[40] Various rating scales are available.[41] Certain portals of entry (e.g., compound fractures) are associated with poorer prognoses. Tetanus may be particularly severe in narcotics addicts, for unknown reasons.[42] *Generalized tetanus* is the most commonly recognized form and often begins with *risus sardonicus* (increased tone in the orbicularis oris) and *trismus* (lockjaw, or masseter rigidity) (Fig. 244.3). Abdominal rigidity may also be present. Progression of the disease typically occurs in a descending pattern. The *generalized spasm* resembles decorticate posturing and consists of opisthotonic posturing with flexion of the arms and extension of the legs (Fig. 244.4). The patient does not lose consciousness and experiences severe pain during each spasm. The spasms are often triggered by sensory stimuli. During the spasm the upper airway can be obstructed, or the diaphragm may participate in the general muscular contraction. Either of these compromises respiration, and even the first such spasm may be fatal. In the modern era of intensive care, however, the respiratory problems are easily managed, and autonomic dysfunction, usually occurring after several days of symptoms, has emerged as the leading cause of death.[43] Symptoms of autonomic hyperactivity, such as hypertension, tachycardia, and hyperthermia, may also be present.

The illness can progress for about 2 weeks, reflecting the time required to complete the transport of toxin, which is already intraaxonal when antitoxin treatment is given. The severity of illness may be decreased by partial immunity.[44] Recovery takes an additional month and is complete unless complications supervene. Rare cases have been described lasting several months.[45] Lower motor neuron dysfunction may not be apparent until spasms remit, and recovery from this deficit in neuromuscular transmission may take additional weeks.[46] Recurrent tetanus may occur if the patient does not receive active immunization because the amount of toxin produced is inadequate to induce immunity.[47] *Localized tetanus* involves rigidity of the muscles associated with the site of spore inoculation. This may be mild and persistent and often resolves spontaneously. Lower motor neuron dysfunction (weakness and diminished muscle tone) is often present in the most involved muscle. This chronic form of the disease probably reflects partial immunity to tetanospasmin.[48] Localized tetanus is more commonly a prodrome of generalized tetanus, however, which occurs when enough toxin gains access to the CNS. *Cephalic tetanus* is a special form of localized disease affecting the cranial nerve musculature, almost always after an apparent head wound (Fig. 244.5). Although earlier reports linked cephalic tetanus to a poor prognosis, more recent studies have revealed many milder cases. A lower motor neuron lesion, frequently producing facial nerve weakness, is often apparent.[49] Extraocular muscle involvement is occasionally noted. *Neonatal tetanus* (Fig. 244.6) follows infection of the umbilical stump, most commonly caused by a failure of aseptic technique if mothers are inadequately immunized.[50] Cultural practices may contribute.[51] The condition usually presents with generalized weakness and failure to nurse; rigidity and spasms occur later. The

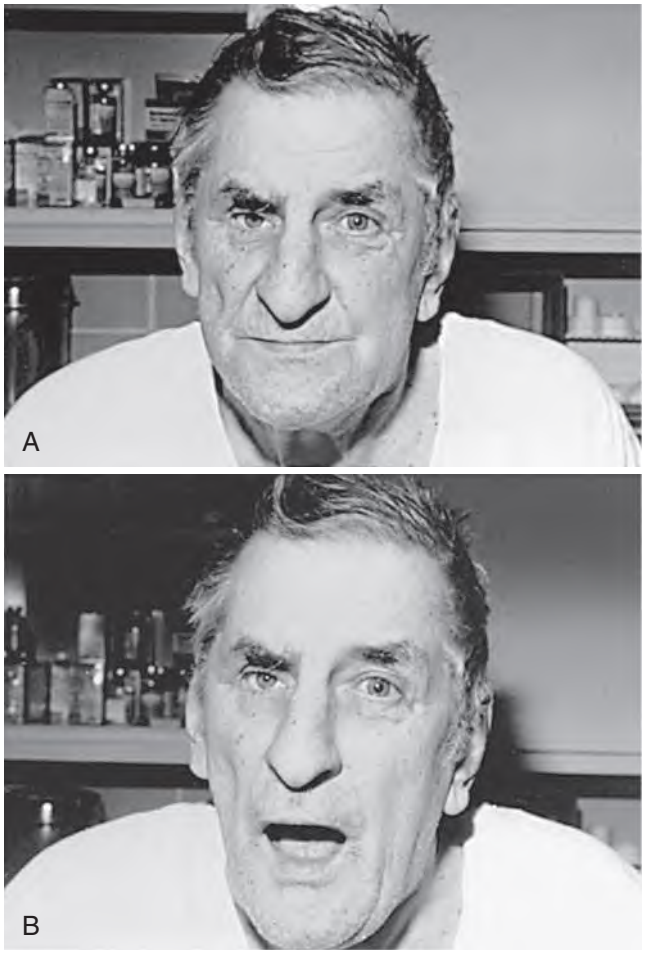

FIG. 244.3 Risus sardonicus and trismus. (A) Risus sardonicus. Note the straightened upper lip at rest. (B) Trismus. The patient is opening his mouth as fully as possible. *(From Bleck TP. Tetanus. In: Scheld WM, Whitley RJ, Durack DT, eds.* Infections of the Central Nervous System. *New York: Raven Press; 1991:603–624.)*

FIG. 244.4 Opisthotonus. *(From Bell C.* Essays on the Anatomy and Physiology of Expression. *2nd ed. London: J. Murray; 1824.)*

FIG. 244.5 Cephalic tetanus. Right facial paresis is present in addition to the grimace. *(From Veronesi R, Focaccia R. The clinical picture. In: Veronesi R, ed.* Tetanus: Important New Concepts. *Amsterdam: Excerpta Medica; 1981:183–206.)*

FIG. 244.6 Neonatal tetanus. *(From Veronesi R, Focaccia R. The clinical picture. In: Veronesi R, ed.* Tetanus: Important New Concepts. *Amsterdam: Excerpta Medica; 1981:183–206.)*

mortality rate exceeds 90%, and developmental delays are common among survivors.[52] Poor prognostic factors include age younger than 10 days, symptoms for fewer than 5 days before presentation to hospital, and presence of risus sardonicus or fever.[53] Apnea is the leading cause of death among neonatal tetanus patients in the first week of life, and sepsis in the second week.[54] Bacterial infection of the umbilical stump leads to sepsis in almost half of infants with neonatal tetanus, which contributes to the substantial mortality despite treatment.[55]

DIAGNOSIS

Tetanus is diagnosed by clinical observation and has a limited differential diagnosis. Laboratory testing cannot confirm or exclude the condition and is primarily useful for excluding intoxications that may mimic tetanus. Electromyographic studies are occasionally useful in questionable cases. Such testing becomes more important when no portal of entry is apparent. Antitetanus antibodies are undetectable in most tetanus patients, but many reports document the disease in patients with antibody levels above the commonly cited "protective" concentration of 0.01 IU/L.[56] Rarely, patients apparently develop antibodies that are not protective.[57]

Attempts to culture *C. tetani* from wounds are not useful in diagnosis because (1) even carefully performed anaerobic cultures are frequently negative; (2) a positive culture does not indicate whether the organism contains the toxin-producing plasmid; and (3) a positive culture may be present without disease in patients with adequate immunity.[58]

Strychnine poisoning, in which glycine is antagonized, is the only condition that truly mimics tetanus; toxicologic studies of serum and urine should be performed when tetanus is suspected, and tetanus should be considered even if strychnine poisoning appears likely. Because the initial treatments of tetanus and strychnine intoxication are similar, therapy is instituted before the assay results are available. Dystonic reactions to neuroleptic drugs or other central dopamine antagonists may be confused with the neck stiffness of tetanus, but the posture of patients with dystonic reactions almost always involves lateral head turning, which is rare in tetanus. Treatment with anticholinergic agents (benztropine or diphenhydramine) is rapidly effective against dystonic reactions. Dental infections may produce trismus and should be sought, but they do not cause the other manifestations of tetanus.

TREATMENT

The patient with tetanus requires simultaneous attention to several concerns. Attention to the airway and to ventilation is paramount at the time of presentation, but the other aspects of care, especially passive immunization, must be pursued as soon as the respiratory system is secure. Table 244.1 presents a suggested management protocol.

Stabilization

Tetanic spasms sometimes demand that the airway be secured before other lines of therapy are possible. An orotracheal tube can be passed under sedation and neuromuscular junction blockade; a feeding tube should be placed at the same time. Because the endotracheal tube may stimulate spasms, an early tracheostomy is usually beneficial.[59]

Management of Muscle Spasms

Benzodiazepines have emerged as the mainstay of symptomatic therapy for tetanus.[60] These drugs are $GABA_A$ agonists and thereby indirectly antagonize the effect of the toxin. They do not restore glycinergic inhibition. The patient should be kept free of spasms and may benefit from the amnestic effects of the drugs as well. Diazepam has been studied most intensively, but lorazepam and midazolam appear equally effective. Tetanus patients have unusually high tolerance for the sedating effect of these agents and commonly remain alert at doses normally expected to produce anesthesia.[61]

The intravenous (IV) formulations of both diazepam and lorazepam contain propylene glycol. At the doses required to control generalized tetanus, this vehicle may produce lactic acidosis.[62] Nasogastric delivery of these agents is often possible, but some tetanus patients develop GI motility disorders and do not absorb drugs well. IV midazolam (5–15 mg/h or more) is effective and does not contain propylene glycol, but it must be given as a continuous infusion because of its short half-life.[63] Propofol infusion is also effective,[64] but the amount necessary to control symptoms may produce the propofol infusion syndrome, characterized by metabolic acidosis, rhabdomyolysis, and cardiac arrhythmias, or exceed the patient's tolerance of the lipid vehicle. When the symptoms of tetanus subside, these agents must be tapered over at least 2 weeks to prevent withdrawal symptoms. Intrathecal baclofen, a $GABA_B$ agonist, is also effective in controlling tetanus but has no clear advantage over benzodiazepines. Neuroleptic agents and barbiturates, previously used for tetanus, are inferior for this indication and should not be used except in settings where benzodiazepines are unavailable. Magnesium sulfate infusion may reduce the need for additional medications to control muscle spasms and cardiovascular instability but does not appear to reduce the need for mechanical ventilation.[65]

Rarely, patients cannot be adequately controlled with benzodiazepines alone. Neuromuscular junction blockade is indicated in such patients,

TABLE 244.1 Suggested Management Protocol for Generalized Tetanus

I. Diagnosis and Stabilization: First Hour After Presentation

A. Assess airway and ventilation. If necessary, perform endotracheal intubation using benzodiazepine sedation and neuromuscular blockade (e.g., vecuronium, 0.1 mg/kg).
B. Obtain samples for antitoxin level, strychnine and dopamine antagonist assays, electrolytes, blood urea nitrogen, creatinine, creatine kinase, and urinary myoglobin determination.
C. Determine the portal of entry, incubation period, period of onset, and immunization history.
D. Administer benztropine (1–2 mg, IV) or diphenhydramine (50 mg, IV) to rule out a dystonic reaction to a dopamine-blocking agent.
E. Administer a benzodiazepine IV (diazepam in 5-mg increments, or lorazepam in 2-mg increments) to control spasm and decrease rigidity. Initially, use a dose that is adequate to produce sedation and minimize reflex spasms. If this dose compromises the airway or ventilation, intubate using a short-acting neuromuscular-blocking agent. Transfer the patient to a quiet, darkened area of the intensive care unit.

II. Early Management Phase: First 24 Hours

A. Administer human tetanus immunoglobulin, 500 U, IM; as an alternative, consider IV pooled immune globulin.
B. At a different site, administer IM adsorbed tetanus toxoid such as tetanus-diphtheria vaccine (0.5 mL) or diphtheria-pertussis-tetanus vaccine (0.5 mL), as appropriate for age. Adsorbed tetanus toxoid without diphtheria toxoid is available for patients with a history of reaction to diphtheria toxoid; otherwise, the correct combination for the patient's age should be used.
C. Begin metronidazole, 500 mg, IV, q6h, for 7–10 days.
D. Perform a tracheostomy after placement of an endotracheal tube and under neuromuscular blockade if spasms produce any degree of airway compromise.
E. Débride any wounds as indicated for their management.
F. Place a soft, small-bore nasal feeding tube or a central venous hyperalimentation catheter, and begin feeding. Patients receiving total parenteral nutrition should also be given parenteral histamine-2 blockade or other gastric protection.
G. Administer benzodiazepines as required to control spasms and produce sedation. Consider magnesium sulfate infusion. If adequate control is not achieved, institute long-term neuromuscular blockade (e.g., vecuronium, 0.8–1.7 μg/kg/min); continue benzodiazepines for sedation with intermittent electroencephalographic monitoring to ensure somnolence. Neuromuscular junction blockade should be discontinued daily to assess the patient's physical examination and to decrease the possibility of excessive accumulation of the blocking agent.

III. Intermediate Management Phase: Next 2–3 Weeks

A. Treat sympathetic hyperactivity with labetalol (0.5–2 mg/min) as needed for blood pressure control) or morphine (0.5–1 mg/kg/h by continuous infusion). Magnesium sulfate infusion may be considered, as well as epidural blockade with a local anesthetic. Avoid diuretics for blood pressure control because volume depletion will worsen autonomic instability.
B. If hypotension is present, initiate saline resuscitation. Place a pulmonary artery catheter and an arterial line, and administer fluids, dopamine, or norepinephrine as indicated.
C. Sustained bradycardia usually requires a pacemaker. Atropine or isoproterenol may be useful during pacemaker placement.
D. Begin prophylactic heparin.
E. Use a flotation bed, if possible, to prevent skin breakdown and peroneal nerve palsies. Otherwise, ensure frequent turning and use antirotation boots.
F. Maintain benzodiazepines until neuromuscular blockade, if used, has been terminated, and the severity of spasms has diminished substantially. Then taper the benzodiazepine dose over 14–21 days.
G. Begin rehabilitation planning.

IV. Convalescent Stage: 2–6 Weeks

A. When spasms are no longer present, begin physical therapy. Many patients require supportive psychotherapy.
B. Before discharge, administer another dose of tetanus-diphtheria vaccine or diphtheria-pertussis-tetanus vaccine.
C. Schedule a third dose of toxoid to be given 4 wk after the second.

IM, Intramuscularly; *IV,* intravenously.
Modified from Bleck TP, Brauner JS. Tetanus. In: Scheld WM, Whitley RJ, Marra CM, eds. Infections of the Central Nervous System. *3rd ed. New York: Lippincott Williams & Wilkins; 2004:625–648.*

with the caveat that sedation is still required for psychological reasons. All the available drugs have side effects, including the potential for prolonged effect after the drug is discontinued. Vecuronium or cisatracurium (by continuous infusion) and pancuronium (by intermittent injection) are adequate choices. These agents should be stopped at least once daily to assess the patient's progress and to observe for possible complications. Electroencephalographic monitoring is a useful adjunct for this purpose.[66]

Wound Management

Most tetanus patients still have the portal of entry apparent when they present. If the wound requires surgical attention, this may be performed after spasms are controlled. However, the course of tetanus is not affected by wound débridement.

Passive Immunization

Passive immunization with human tetanus immune globulin (HTIG) shortens the course of tetanus and may lessen its severity. A dose of 500 units appears as effective as larger doses.[67] A meta-analysis of the benefits of intrathecal HTIG therapy was inconclusive.[68] However, in a randomized trial, the administration of intrathecal HTIG with intramuscular HTIG resulted in shorter duration of spasms, shorter hospital stay, and decreased respiratory assistance demands compared with intramuscular HTIG alone.[69] Pooled IV immune globulin has been proposed as an alternative to HTIG,[70] although this should be approached with caution.[71] Active immunization must also be initiated.

Antimicrobial Therapy

The role of antimicrobial therapy in tetanus remains debated. The in vitro susceptibilities of *C. tetani* include metronidazole, penicillins, cephalosporins, imipenem, macrolides, and tetracycline. A study comparing oral metronidazole to intramuscular penicillin showed better survival, shorter hospitalization, and less progression of disease in the metronidazole group.[72] This may reflect a true advantage of metronidazole over penicillin, but it more likely corresponds to a negative effect of penicillin, a potent $GABA_A$ antagonist. Topical antibiotic application to the umbilical stump appears to reduce the risk for neonatal tetanus.[7]

Management of Autonomic Dysfunction

Autonomic dysfunction generally reflects excessive catecholamine release and may respond to combined α-adrenergic and β-adrenergic blockade with IV labetalol.[73] β-Blockade alone is rarely used because the resulting unopposed α effect may produce severe hypertension. If β-blockade is chosen, the short-acting agent esmolol should be used.[74] Other approaches to hypertension include morphine infusion,[75] magnesium sulfate infusion,[76] and epidural blockade of the renal nerves.[77] Hypotension is less common but, if present, may require norepinephrine infusion. Myocardial dysfunction is also common[78] and may represent a further reflection of catecholamine excess.[79]

Nutritional Support

Nutritional support should be started as soon as the patient is stable. The volume of enteral feeding needed to meet the exceptionally high caloric and protein requirements of these patients may exceed the capacity of the GI system.

The mortality rate in mild and moderate tetanus at present is about 6%; for severe tetanus, it may reach as high as 60%, even in expert centers.[80] A well-designed protocol for the critical care of tetanus patients can substantially reduce morbidity and mortality.[81] Among adults, age has little effect on mortality, with octogenarians and nonagenarians faring as well as middle-aged patients.[82] Tetanus survivors often have serious psychological problems related to the disease and its treatment that persist after recovery and may require psychotherapy.[83]

Prevention

Tetanus is preventable in almost all patients, leading to its description as the "inexcusable disease."[84] Tetanus toxoid (TT), a heat-inactivated toxin, was developed in 1924.[85] The vaccine was initially used among military personnel in World War II. As a result, tetanus accounted for

only 12 of nearly 3 million hospitalizations during the war; five cases were fatal.[86,87]

The Advisory Committee on Immunization Practices (ACIP) recommends a primary tetanus vaccination in combination with diphtheria and pertussis (DTaP) at 2, 4, 6, and 12 to 18 months, and at 4 to 6 years.[88] In 2005 the US Food and Drug Administration approved the use of a new vaccine formulation: tetanus, reduced diphtheria toxoid, and acellular pertussis (Tdap).[89] Tdap should be administered once to all adolescents at age 11 to 18 years (also see Chapter 316).[88]

Serologic analysis of the US population suggests that tetanus immunity wanes with age.[90,91] Although 80% of patients aged 6 to 39 years were noted to have protective antibodies to tetanus, only 28% of patients older than 70 years were seropositive.[90] Therefore it is recommended that all adults receive a tetanus diphtheria toxoid (Td) booster every 10 years. Between the ages of 19 and 64 years, one of these Td boosters should be replaced with a single dose of Tdap. Tdap was previously recommended only for adults older than 65 years who had not previously received Tdap and who anticipated close contact with infants aged younger than 1 year; however, in 2012 ACIP recommended Tdap for all adults aged 65 years and older.[92] That same year, ACIP also made a recommendation that all pregnant women receive Tdap during each pregnancy, regardless of prior vaccination status.[93] All patients who seek medical attention for a wound should have their tetanus immunization history reviewed. Patients with minor, clean wounds who have not received at least three doses of a tetanus toxoid–containing vaccine or whose last dose was 10 years or more prior should receive a tetanus toxoid–containing vaccine; which vaccine is administered depends on patient's age and vaccine history. Patients with more serious or contaminated wounds who have an incomplete or unknown tetanus vaccine history should receive a tetanus toxoid–containing vaccine and HTIG. HTIG binds directly to toxin, providing temporary immunity. HTIG should be given at a different site and with a different syringe than vaccine. Patients with serious wounds who have previously received at least three doses of a tetanus toxoid–containing vaccine but whose last dose was 5 or more years ago should receive a tetanus booster without HTIG.[94] Tetanus-prone wounds are characterized by devitalized tissue, such as a crush injury, or by a wound with potential contamination with dirt or rust.

Some patients with humoral immune deficiencies may not respond adequately to toxoid injection[95]; such patients should receive passive immunization for tetanus-prone injuries regardless of the period since the last booster. About half of patients lose tetanus immunity after chemotherapy for leukemia or lymphoma.[96] Patients who have undergone bone marrow or stem cell transplantation require revaccination after the procedure.[97] It is recommended that three Td boosters be given, the first of which should be administered 6 to 12 months after transplantation. One of the three Td doses should be replaced with Tdap.[98] Antibody production by the transplanted immune cells may play a minor role in subsequent host immunity.[99] Most young patients with human immunodeficiency virus (HIV) infection appear to retain antitetanus antibody production if their primary immunization series was completed before they acquired HIV[100]; however, only a minority respond adequately to booster immunization.[101] Vitamin A deficiency interferes with the response to tetanus toxoid.[102] There have been rare reports of patients with protective antitoxin antibodies in their serum from immunization developing tetanus as well.

Neonatal tetanus may occur because of inadequate immunization of the mother. Although a full series of maternal immunizations is ideal, even one or two doses of tetanus toxoid confer substantial protection against neonatal tetanus.[103] Application of topical antimicrobial agents to the umbilical cord stump markedly decreases the incidence of neonatal tetanus when maternal immunization is insufficient.[104] Mild reactions to tetanus toxoid (e.g., local tenderness, edema, low-grade fever) are common. More severe reactions are rare and likely are due to a hypersensitivity response to the preservative thimerosal.[105] Although there have been reports suggesting a connection between tetanus immunization and Guillain-Barré syndrome, a careful epidemiologic analysis did not confirm an association.[106]

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245 Botulism (*Clostridium botulinum*)

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SHORT VIEW SUMMARY

Definition
- Botulism is a toxin-mediated paralytic illness caused by *Clostridium botulinum.* It is classified as foodborne botulism, infant botulism, wound botulism, iatrogenic botulism, botulism of undetermined etiology, or inhalational botulism.

Epidemiology
- Foodborne botulism occurs in outbreaks, whereas other forms are sporadic.
- Foodborne botulism is associated with home-canned or fermented foods.
- Infant botulism historically is associated with honey ingestion.
- Wound botulism is associated with injection drug use of "black-tar" heroin.
- Botulinum toxins A and B are used for therapeutic and cosmetic purposes and may cause iatrogenic botulism.
- Botulism is a potential bioterrorism agent deployed by aerosol or ingestion.

Microbiology
- *C. botulinum* is a gram-positive, strictly anaerobic bacillus that forms a subterminal spore.
- *C. botulinum* produces seven distinct toxins, designated types A through G.

Diagnosis
- Presumptive diagnosis is based on clinical presentation: acute, bilateral cranial neuropathies with symmetrical descending weakness.
- Mouse bioassay is the gold standard for botulinum toxin.
- Culture of serum, stool, and environmental samples requires strict anaerobic conditions and is low yield.
- Characteristic electrophysiologic study findings are suggestive of botulism.

Therapy
- Supportive care remains the mainstay of botulism treatment.
- Heptavalent botulinum antitoxin is available for noninfant botulism in the United States.
- Human botulinum immune globulin (BabyBIG) is available for the treatment of infant (younger than 1 year) botulism. To obtain it, contact the California Department of Health Services, Infant Botulism Treatment and Prevention Program (510-231-7600; www.infantbotulism.org).

Prevention
- Proper food preparation prevents foodborne botulism.
- There is no currently available vaccine.

Botulism and tetanus result from intoxication with the protein neurotoxins elaborated by two related species of *Clostridium.* The toxins are very similar in structure and function but differ dramatically in their clinical effects because they target different cells in the nervous system. Botulinum neurotoxins predominantly affect the peripheral neuromuscular junction and autonomic synapses and primarily manifest as weakness. In contrast, although tetanus toxin can affect the same systems, its effects reflect tropism for inhibitory cells of the central nervous system (CNS) and primarily manifest as rigidity and spasm. Both conditions have potentially high fatality rates, and both are preventable through education and public health measures.

Clostridium botulinum produces most cases of botulism, with a few other clostridial strains accounting for the remainder. Botulinum toxins are designated types A through G based on antigenic differences.[1] Type C/D hybrid toxins have also been described.[2] Types A, B, E, and F produce human disease, whereas types C and D are almost exclusively confined to animals.[3] Type G toxin has not been associated with naturally acquired disease. In 2014, a possible new toxin was discovered as a result of a case of infant botulism in which the toxin produced could not initially be neutralized by any of the existing anti-A through anti-G antitoxins in mouse bioassay.[4] There remains some uncertainty regarding whether this toxin should be classified as a distinct toxin. Although it was originally designated as a distinct toxin labeled as type H, the Centers for Disease Control and Prevention (CDC) has used the term "F/A Hybrid" because it can be neutralized by existing serotype A antitoxin, albeit at higher doses.[5–7] For this reason, most do not consider this to be a distinct toxin. A novel monoclonal antibody directed against this toxin has been described but is not commercially available.[8] In addition, a novel toxin has recently been identified through genomic sequencing of a *C. botulinum* strain, although the clinical implications of this finding are unclear.[9] The clinical forms of botulism include *foodborne botulism, infant botulism, wound botulism*, and *botulism of undetermined etiology.* Botulinum A toxin has achieved prominence as a therapeutic modality in conditions that result from excessive muscle activity (e.g., torticollis), leading to rare cases of *iatrogenic botulism.* Botulinum toxin has also been developed as a weapon, which could be used to contaminate food or beverage supplies or could be aerosolized.

HISTORY OF BOTULISM

The term *botulism* derives from the Latin word *botulus,* or sausage. Outbreaks of poisoning related to sausages and other prepared foods occurred in Europe in the 19th century. Justinus Kerner, a district health officer in southern Germany, recognized the connection between sausage and the paralytic illnesses of 230 patients in 1820 and made sausage poisoning a reportable disease.[10] At about the same time, physicians in Russia recognized a disease with similar symptoms, which they termed *fish poisoning.*[11] In 1897, van Ermengen published the first description of *C. botulinum* and showed that the organism elaborated a toxin that could induce weakness in animals.[12] This was subsequently shown to be type A toxin; type B was discovered in 1904.[13] Wound botulism was described in 1943,[14] and infant botulism in 1976.[15] The occurrence of sporadic cases without an apparent etiology, many related to gastrointestinal colonization, was first reported in 1986.[16] Type A toxin was isolated and purified in 1946.[17]

EPIDEMIOLOGY

Foodborne botulism is most frequently recognized in outbreaks, whereas the other forms are sporadic. Although commercially canned foods were commonly the source of toxin in the early part of this century, home-canned vegetables, fruits, and fish products are now the most common sources. In some cultures, such as among Alaskan Natives, preferred food preparation practices involving fish fermentation commonly lead to botulism.[18] In China, homemade fermented beans are the leading cause.[19] Commercial foods and restaurants are still occasional sources.[20,21]

Consumption of peyote for religious reasons has resulted in botulism.[22] Pruno, an illicit, prison-brewed alcoholic beverage, has also emerged as a cause of botulism.[23] In the United States, 263 cases occurred from 1990 to 2000 because of 163 foodborne botulism events (17–43 cases per year).[24] In 2015 there were 39 cases of confirmed foodborne botulism in the United States, primarily associated with five outbreaks.[25]

Infant botulism primarily occurs with toxin types A, B, or F. In the past, infections were attributed to honey ingestion,[26] but other sources have emerged as feeding honey to infants has been discouraged.[27] In the absence of competing microbiota found in children and adults, *C. botulinum* colonizes the intestine of infants (ages 6 days to 12 months). Infection occurs as a consequence of absorption of toxin produced by *C. botulinum* in situ.[28] From 1992 to 2006, 2419 cases of infant botulism were identified in the United States (average, 2.1 cases per 100,000 live births).[29] Two infants without other exposures are believed to have contracted botulism through soil contamination.[30] Rare cases of infant botulism have been associated with *Clostridium baratii*[31] or *Clostridium butyricum*.[32]

Wound botulism may be caused by either type A or type B organisms. In such cases, *C. botulinum* spores contaminate the wound, leading to subsequent germination and toxin production. Almost exclusively associated with injection drug use of "black-tar" heroin, wound botulism was first reported in the United States in the 1990s. Spore contamination of heroin during preparation can lead to infection, particularly in patients who inject by "skin-popping" (i.e., drug injection into tissue rather than the vein).[33,34] The majority of wound botulism cases have been reported in California. However, black-tar heroin–related cases have also been described in Europe, including 12 cases in Germany in 2005.[35–37]

Adult botulism of unknown etiology usually involves type A toxin, but types B and F have also been implicated.[38] Affected adults become colonized with and subsequently infected by toxin-producing clostridia. Adults at risk include those with loss of bowel microbiota because of anatomic abnormalities, functional disorders, or antibiotic use.[39,40–42] In the setting of adult botulism of unknown etiology attributed to type F the disease was caused by *C. baratii*.[42]

Botulinum toxin types A and B are approved by the US Food and Drug Administration (FDA) for cosmetic and therapeutic purposes (e.g., blepharospasm, strabismus, cervical dystonia). Iatrogenic botulism cases are uncommon but have been reported with the therapeutic[43] and unlicensed cosmetic use of botulinum toxin A.[44,45]

Rare cases of inhalational botulism have been associated with the intranasal use of contaminated cocaine.[46] Inhalation is also one of the potential routes of a bioterrorist attack with botulinum toxin. *C. botulinum* has been considered a high enough probability for use in bioterrorism that it has been targeted by a blue ribbon panel for special research emphasis. A bioterrorist attack with this toxin could cause intoxication via ingestion or as an aerosol. In the event of intentional contamination of food with botulinum toxin, the signs and symptoms of the victims of such an attack would be indistinguishable from a natural outbreak of botulism, except that epidemiologic investigation might reveal that the common food ingested was not typically associated with botulism or that different foods in the same area were all contaminated. Introduction of toxin into milk trucks or other large, closed food or beverage transports would produce sporadic cases. In such a circumstance, individual clinicians would be unlikely to recognize an attack early in its development. Automated systems for the collection of epidemiologic data are required for this purpose.[47]

Predicting the consequences of dissemination into the environment is more problematic because there are no data regarding the stability of the toxin in water or sunlight. One CDC expert estimated that an aerosol release of toxin could affect 10% of people within 500 meters.[48] Once in the atmosphere, the decay rate of the toxin is estimated to be 1% to 4% per minute. Modeling an aerosol exposure suggests that substantial inactivation could take up to 2 days but would be accelerated by extremes of temperature and humidity.[48]

MICROBIOLOGY OF *CLOSTRIDIUM BOTULINUM*

C. botulinum is a large, gram-positive, strictly anaerobic bacillus that forms a subterminal spore.[49] The species is divided into four physiologic groups. Group I organisms are proteolytic in culture and can produce toxin types A, B, and F. Group II organisms are nonproteolytic and can produce toxin types B, E, and F. Group III organisms produce toxin types C and D, and group IV produces type G. A single strain almost always produces only one toxin type. Group II organisms grow optimally between 25°C and 30°C, and the other groups grow best between 30°C and 37°C. Although each strain of the organism typically contains several plasmids, only type G toxin is encoded on a plasmid (unlike *Clostridium tetani,* in which the toxin apparently is always encoded on a plasmid).[50]

C. botulinum spores are found throughout the world in soil samples and marine sediments.[51] These spores are able to tolerate 100°C at 1 atm for several hours; because boiling renders solutions more anaerobic, it may actually favor the growth of *C. botulinum*.[52] Proper preparation of food in a pressure cooker will render spores inert.

PATHOGENESIS

In foodborne botulism, toxin is ingested with the food in which it was produced. It is absorbed primarily in the duodenum and jejunum and passes into the bloodstream, by which it reaches peripheral cholinergic synapses (including the neuromuscular junction). Infant botulism and probably adult botulism of unknown etiology have a somewhat different pathogenesis in that they are acquired through the ingestion of spores rather than preformed toxin. The infant's intestinal microbiota is thought to be particularly permissive for the germination of spores, which leads to the production of toxin. The spores are acquired from environmental sources contaminated with soil in which botulinum spore counts are high.[53] In adults, achlorhydria and antibiotic use may predispose to gastrointestinal colonization with *C. botulinum*. In cases of wound botulism, spores are introduced into a wound, where they germinate and produce toxin. Lastly, in inhalational botulism, the toxin crosses through the pulmonary alveolar epithelium to gain access to the bloodstream.[54] The clinical manifestations of botulism depend on the type of toxin produced, rather than the site of its production.

Botulinum toxin is synthesized as a single polypeptide chain of low potency; the molecular weight varies from 150 to 165 kDa, depending on the toxin type. The botulinum toxins are zinc-dependent metalloproteinases,[55] as is tetanospasmin (the neurotoxin associated with *C. tetani)*. The toxin is then nicked by a bacterial protease to produce two chains, with the light chain constituting about one-third of the total mass. As with tetanospasmin, the chains remain connected by a disulfide bond. The nicked toxin type A becomes, on the basis of molecular weight, the most potent toxin found in nature. In contrast to the spores, the toxin is heat labile. Different toxin types may undergo different postsynthetic processing.[56]

In the laboratory, the clostridial toxins have provided a major tool for understanding the mechanisms of neurotransmitter release. Once present at the synapse, the toxins prevent the release of acetylcholine (ACh). This appears to result from a three-stage process.[57] The heavy chain of the toxin mediates binding to presynaptic receptors. The nature of these receptors is uncertain; different toxin types bind to different receptors, with type B receptors outnumbering type A receptors by a factor of four.[58] The toxin enters the cell by receptor-mediated endocytosis.[59] Once inside the neuron, the toxin types differ in the mechanisms by which they inhibit ACh release.[60] The release of synaptic vesicles by an action potential is initiated by an abrupt rise in the intracellular free Ca^{2+} concentration, mediated by voltage-dependent calcium channels (Fig. 245.1).[61] This increase in free calcium triggers an interaction between synaptotagmin (in the vesicle membrane) and syntaxin (on the presynaptic cell membrane), clamping the vesicle to the presynaptic membrane. Synaptobrevin (also referred to as *vesicle-associated membrane protein*[62]) also binds to syntaxin and appears to dock the vesicle to the membrane at the proper location for fusion. There are different isoforms of synaptobrevin within neurons; a protein termed *cellubrevin* performs a similar function in nonneuronal secretory cells.[63] Synaptophysin, the third major component of this mechanism, probably forms the fusion pore that allows release of the vesicle contents into the synaptic cleft.[64]

Clostridial neurotoxins inhibit vesicle release by cleaving peptide bonds in these proteins.[65] Each toxin has a specific locus of activity. Tetanospasmin, along with botulinum neurotoxins B, D, F, and G, cleaves synaptobrevin.[66,67] Tetanospasmin and botulinum neurotoxin B appear

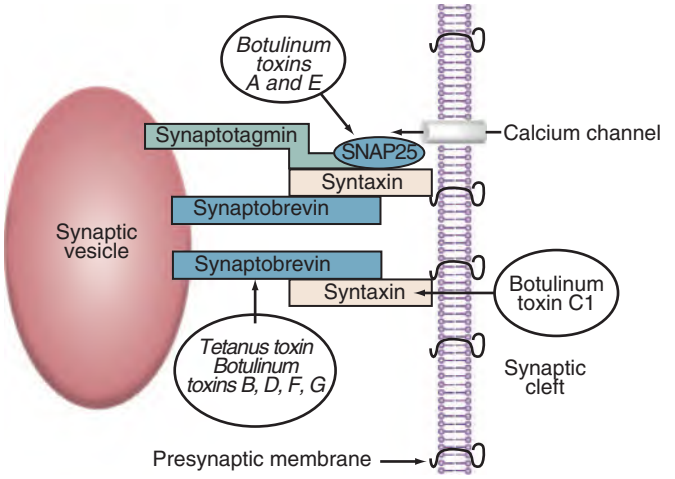

FIG. 245.1 Components of the transmitter release mechanism. SNAP25, synaptosomal-associated protein-25. *(From Bleck TP, Brauner JS. Tetanus. In: Scheid WM, Whitley RJ, Durack DT, eds.* Infections of the Central Nervous System. *2nd ed. New York: Raven Press; 1997:629–653.)*

to share the same cleavage site on synaptobrevin.[68] In contrast, botulinum toxins A[69] and E act on the 25-kDa synaptosomal-associated protein 25 (SNAP25),[70] and botulinum toxin C1 affects syntaxin. The toxins affect only the free proteins; once they have complexed to cause transmitter release, they are not subject to attack.[71] Synaptobrevin and synaptotagmin cleavage also occurs normally, as an effect of an endogenous protease, and these proteins are probably involved in organelle recycling.[72] The endogenous protease does not appear homologous to the clostridial toxins. However, the result is that stimulation of the presynaptic cell (e.g., the alpha motor neuron) fails to produce transmitter release, thus producing paralysis in the motor system or autonomic dysfunction when parasympathetic nerve terminals or autonomic ganglia are involved.

Once damaged, the synapse was originally thought to be rendered useless. Because of the widespread interest in the therapeutic use of botulinum toxin, substantial research into the mechanisms of recovery is underway. The initial recovery of function in type A botulism requires sprouting of the presynaptic axon and the subsequent formation of a new synapse. Later, the original synapse recovers, and the newer ones are pruned away.[73] With type F botulism, recovery is substantially faster, suggesting that the original synapse regains function more rapidly.[74]

Botulinum toxin is transported within nerves in a manner analogous to tetanospasmin and can thereby gain access to the CNS. However, symptomatic CNS involvement is rare.[75]

CLINICAL MANIFESTATIONS

The classic presentation of botulism is that of a patient who develops acute, bilateral cranial neuropathies associated with symmetrical descending weakness. The CDC suggests attention to these cardinal features: (1) fever is absent (unless a complicating infection occurs), (2) the neurologic manifestations are symmetrical, (3) the patient remains responsive, (4) the heart rate is normal or slow in the absence of hypotension, and (5) sensory deficits do not occur (except for blurred vision).[42] The first two features were important for the exclusion of poliomyelitis. Atypical findings such as unilateral symptoms or ascending paralysis have been described in up to 7% of reported cases.[76]

Foodborne botulism usually develops 12 to 36 hours after toxin ingestion. The patient initially reports nausea and a dry mouth, and diarrhea may occur at this stage. If diarrhea does occur, it is due to other substances in the ingested material; botulinum toxin itself will produce constipation when it affects parasympathetic autonomic ganglia. Evidence of cranial nerve dysfunction most commonly starts with the eyes, reflecting parasympathetic involvement (blurred vision as a result of pupillary dilation) or involvement of cranial nerves III, IV, or VI.[77] Pupillary reactions may remain abnormal for months after motor recovery.[78] Nystagmus is occasionally noted, usually in type A disease. Lower cranial nerve dysfunction manifests as dysphagia, dysarthria, and hypoglossal weakness. Weakness then spreads to the upper extremities, the trunk, and the lower extremities. Respiratory dysfunction may result from either upper airway obstruction (the weakened glottis tends to close during attempted inspiration) or diaphragmatic weakness. Patients who need mechanical ventilation require mean periods of 58 days (type A) and 26 days (type B) for ventilatory weaning.[79] Recovery may not begin for up to 100 days.[80] Autonomic problems may include gastrointestinal dysfunction, alterations in resting heart rate, loss of responsiveness to hypotension or postural change, hypothermia, and urinary retention.[81]

Hughes summarized published reports to analyze differences in the clinical findings of intoxication with different toxin types (Table 245.1).[82] Type A is significantly more commonly associated with dysarthria, blurred vision, dyspnea, diarrhea, sore throat, dizziness, ptosis, ophthalmoplegia, facial paresis, and upper extremity weakness. Types B and E appear to produce more autonomic dysfunction. None of these differences is diagnostic of the toxin type, however. It is important to note that the pupils are either dilated or unreactive in less than 50% of patients; although these are useful signs when present, their absence in no way diminishes the likelihood of botulism.

Infants with botulism present with constipation, which may be followed by feeding difficulties, hypotonia, increased drooling, and a weak cry.[83] Upper airway obstruction may be the initial sign[84] and is the major indication for intubation.[53] In severe cases, the condition progresses to include cranial neuropathies and respiratory weakness, with ventilatory failure occurring in about 50% of diagnosed patients. The condition progresses for 1 to 2 weeks and then stabilizes for another 2 to 3 weeks before recovery starts.[85] Relapses of infant botulism may occur.[86]

Wound botulism lacks the prodromal gastrointestinal disorder of the foodborne form but is otherwise similar in presentation. Fever, if present, reflects wound infection rather than botulism. The wound itself may rarely appear to be healing well while neurologic manifestations are occurring. Conversely, *C. botulinum* infection may produce abscesses[87]; botulism has also been reported as a result of sinusitis with this organism after cocaine inhalation.[88] The reported incubation period varies from 4 to 14 days.

The signs and symptoms exhibited by victims of inhalational botulism are the same as those seen with ingestion. The latency between exposure and clinical disease after inhalation appears to be between 12 hours and 3 days, with maximal disease by about 5 days.[48]

Botulinum toxin has been used to treat a variety of chronic pain syndromes, achalasia, and anal fissures.[89] It has also achieved widespread notoriety for its use in cosmetic procedures. In 2004, four patients developed clinical symptoms of botulism after the unlicensed cosmetic use of botulinum toxin A.[44]

DIAGNOSIS

A history appropriate to the type of botulism suspected is the most important diagnostic test. If others are already affected, the condition is easily recognized. However, because the toxin may not be evenly distributed in foodstuffs, the absence of other patients does not eliminate the diagnosis. A screening tool has been suggested to help facilitate timely diagnosis of botulism, which is intended to aid physicians in identifying patients who may have botulism, although it is not intended for diagnosis. This tool uses three criteria: (1) afebrile status; (2) at least one of the following symptoms: blurred vision, double vision, difficulty speaking, change in sound of voice, dysphagia, or thick tongue; and (3) at least one of the following signs: ptosis, extraocular palsy, facial paralysis, fixed pupils, or descending paralysis. Based on a review of 241 cases, this tool showed a sensitivity of 87%.[90] This tool would likely also pick up many botulism mimics; however, it could facilitate earlier treatment.

Botulism has a limited differential diagnosis. Myasthenia gravis and Lambert-Eaton myasthenic syndrome (LEMS) each share some of the characteristics of botulism, but are rarely fulminant, and myasthenia lacks autonomic features. An edrophonium test may be considered, but an improvement in strength is not pathognomonic of myasthenia gravis and has been reported in botulism[91]; however, edrophonium is currently out of production. Tick paralysis is excluded by a careful physical examination because the *Dermacentor* tick will still be attached.

TABLE 245.1 Symptoms and Signs in Patients With the Common Types of Human Botulism

	TYPE A (%)	TYPE B (%)	TYPE E (%)
Neurologic Signs and Symptoms			
Dysphagia	96	97	82
Dry mouth	83	100	93
Diplopia	90	92	39
Dysarthria	100	69	50
Upper extremity weakness	86	64	NA
Lower extremity weakness	76	64	NA
Blurred vision	100	42	91
Dyspnea	91	34	88
Paresthesias	20	12	NA
Gastrointestinal Signs and Symptoms			
Constipation	73	73	52
Nausea	73	57	84
Vomiting	70	50	96
Abdominal cramps	33	46	NA
Diarrhea	35	8	39
Miscellaneous Symptoms			
Fatigue	92	69	84
Sore throat	75	39	38
Dizziness	86	30	63
Neurologic Findings			
Ptosis	96	55	46
Diminished gag reflex	81	54	NA
Ophthalmoparesis	87	46	NA
Facial paresis	84	48	NA
Tongue weakness	91	31	66
Pupils fixed or dilated	33	56	75
Nystagmus	44	4	NA
Upper extremity weakness	91	62	NA
Lower extremity weakness	82	59	NA
Ataxia	24	13	NA
DTRs diminished or absent	54	29	NA
DTRs hyperactive	12	0	NA
Initial Mental Status			
Alert	88	93	27
Lethargic	4	4	73
Obtunded	8	4	0

DTRs, Deep tendon reflexes; NA, not available.
Data from Tacket CO, Rogawski MA. Botulism. In: Simpson LL, ed. Botulinum Neurotoxin and Tetanus Toxin. *San Diego, CA: Academic Press; 1989:351–378; Hughes JM. Botulism. In: Scheld WM, Whitley RJ, Durack DT, eds.* Infections of the Central Nervous System. *New York: Raven Press; 1991:589–602; and Weber JT, Hibbs RG, Darwish A, et al. A massive outbreak of type E botulism associated with traditional salted fish in Cairo.* J Infect Dis. *1993;167:451–454.*

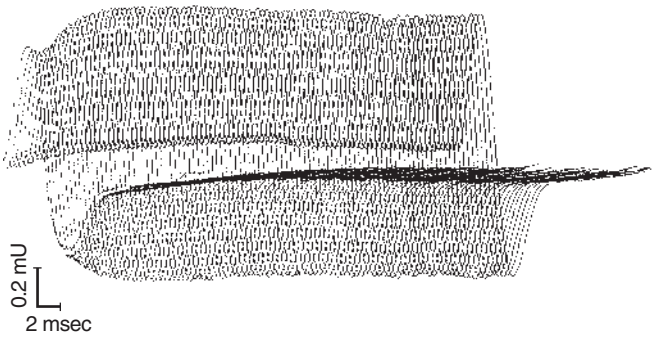

FIG. 245.2 Repetitive nerve stimulation in infant botulism. Note the increment in response amplitude during the initial stimulations. *(Courtesy Vern Juel, MD, Department of Neurology and Laboratory of Electromyography, University of Virginia, Charlottesville, VA.)*

Classic acute inflammatory demyelinating polyneuropathy (Guillain-Barré syndrome) frequently begins with sensory complaints, rapidly produces areflexia, rarely begins with cranial nerve dysfunction, and does not alter pupillary reactivity. Patients with botulism do not become areflexic until the affected muscle group is completely paralyzed. The Miller Fisher variant of Guillain-Barré syndrome presents with oculomotor dysfunction and may produce other cranial neuropathies but includes a prominent ataxia that is lacking in botulism. Patients with polio are febrile on presentation and have asymmetrical weakness. Magnesium intoxication may mimic botulism.[92] Rarely, botulism may be confused with diphtheria, organophosphate poisoning, or brainstem infarction.[93]

Conventional diagnosis of botulism relies on the demonstration of toxin in serum, gastric secretions, stool, or food samples. The most sensitive means of botulism toxin detection has traditionally been the mouse bioassay.[94] After receiving an injection of sample, mice are followed for the development of symptoms. Toxin type may be determined by injecting infected mice with type-specific botulism antitoxin. Botulism symptoms are absent from infected mice that receive the appropriate antitoxin. Confirmation and toxin typing are obtained in almost 75% of cases.[95] The mouse bioassay is labor and resource intensive, and therefore the testing is performed in a limited number of public health laboratories. A novel assay based on mass spectroscopy has been reported to have greater sensitivity than the mouse bioassay and detected botulinum toxin in an infant in whom polymerase chain reaction, bacterial cultures, and mouse bioassay were negative.[96,97] In addition, this assay can be performed in 7 to 8 hours, compared with the mouse bioassay, which may take several days. A Biosafety Level 2 containment facility is a minimum requirement for *C. botulinum* detection and evaluation, given its potency. Testing should be performed under the direction of local state or health departments. If consultation is required after hours, the regional Poison Center (800-222-1222) or the CDC's Emergency Operations Center (770-488-7100) may be contacted.

Anaerobic cultures of serum, stool, and the implicated food, if available, may assist in making the diagnosis. However, samples rarely yield *C. botulinum* because strict anaerobic conditions are required for growth, and competing fecal microbiota or nontoxigenic *C. botulinum* strains can make isolation difficult. Toxin excretion may continue up to 1 month after the onset of illness, and stool cultures may remain positive for a similar period.

Enzyme-linked immunosorbent assay has been used to detect botulinum toxin in clinical specimens and in contaminated food samples, such as fish fillets, canned salmon and corned beef, pasta products, and canned vegetables.[98–101] Techniques based on the polymerase chain reaction and on mass spectrometry are also being explored as potential diagnostic tools.[102,103,104]

Electrophysiologic studies reveal normal nerve conduction velocities; the amplitude of compound muscle action potentials is reduced in 85% of cases, although not all motor units may demonstrate this abnormality.[105] Repetitive nerve stimulation at high rates (20 Hz or greater, compared with the 4-Hz rate used in the diagnosis of myasthenia gravis) may reveal a small increment in the motor response (Fig. 245.2), as opposed

to the decrement expected in myasthenia. This test is very uncomfortable and should not be requested unless botulism or LEMS is a serious consideration. Botulism can be distinguished electrophysiologically from LEMS.[106] In infant botulism, the increments may be very dramatic. In questionable cases, single-fiber electromyography studies may be useful. There is currently some debate regarding the sensitivity of electrodiagnostic techniques in cases of infant botulism.[107] The therapeutic use of botulinum A toxin for dystonic disorders can produce electrophysiologic evidence of toxin dissemination to distant sites.[108]

If botulinum toxin is used as a biologic weapon, the diagnosis would depend on the route of exposure. Contaminated food or beverages would result in an epidemic resembling that of a natural foodborne outbreak. A deliberate release of botulinum toxin should be suspected if patients with acute flaccid paralysis and prominent bulbar palsies present in large numbers. An unusual toxin type (such as C, D, F, G, or F/A Hybrid) or symptoms among patients with a common geographic location may suggest an act of bioterrorism.[48] The amount of inhaled toxin–producing disease would probably not produce measurable toxin in blood or other patient samples, except perhaps for nasopharyngeal secretions. Therefore, current approaches to the diagnosis of botulism and the detection of botulinum toxin would be of limited value during an attack. The diagnosis can be confirmed most rapidly with electromyography.

THERAPY

The importance of supportive therapy for botulism is underlined by the progressive improvement in mortality rates with advances in critical care, especially ventilatory support. The decision to intubate should be based on (1) bedside assessment of upper airway competency and (2) changes in vital capacity. (In general, an appropriately performed vital capacity measurement of <12 mL/kg frequently indicates intubation. However, the facial weakness of botulism may preclude a tight seal on the spirometer mouthpiece, invalidating the test.) One should not wait for the partial pressure of arterial carbon dioxide ($Paco_2$) to rise or the oxygen saturation to fall before intubating the patient. In contrast to tetanus, the autonomic dysfunction of botulism is rarely life-threatening, and patients who receive appropriate airway and ventilator management should recover unless complications supervene. Patients intubated with high-volume, low-pressure endotracheal tubes should not automatically undergo tracheostomy, regardless of the duration of intubation, unless required for mechanical reasons.[109] If contaminated food may still reside in the gastrointestinal tract, purgatives may be useful unless ileus has occurred. The detailed critical care management of botulism patients is beyond the scope of this text; Tacket and Rogawski have presented a useful approach.[52]

In March 2010, the CDC announced the availability of a new, heptavalent botulinum antitoxin (HBAT). HBAT contains equine-derived antibody to the seven known botulinum toxin types (A through G).[110] It replaced the trivalent (types A, B, and E) equine serum and was FDA approved in 2013. It is the only antitoxin available for noninfant botulism in the United States and has been found to be relatively safe and effective in adults, particularly when administered within 2 days of symptom onset,[111] although there is a 1% to 2% risk of anaphylactic reaction.[112] Because of the risk of serious adverse reactions and the potential for lifelong sensitization to equine proteins, HBAT is not approved for administration in infants younger than 1 year.[113] Human botulinum immune globulin (BabyBIG) was approved by the FDA in 2003 for the treatment of infant (younger than 1 year) botulism. Use of BabyBIG has resulted in decreased length of intensive care unit stay, length of mechanical ventilation, and overall length of hospital stay.[114] These benefits are greatest when BabyBIG is administered within the first 3 days of hospitalization.[115] The current recommended dose is 50 mg/kg as an intravenous infusion, although the recommended dose may vary with each manufactured lot and should be confirmed. For suspected cases of infant botulism in any state, the California Department of Health Services, Infant Botulism Treatment and Prevention Program should be contacted (510-231-7600; www.infantbotulism.org). In the event of intentional dissemination of botulinum toxin, heptavalent antitoxin may be dispensed by the Department of Defense.[48]

Patients with wound botulism should also undergo débridement, even if the wound appears to be healing well. Anaerobic cultures should be obtained at the time of surgery. The value of local instillation of antitoxin is unknown. The role of antibiotic treatment is untested, but penicillin G (10–20 million units daily) is frequently recommended. Metronidazole may be an effective alternative. Aminoglycosides and tetracyclines, which can impair neuron calcium entry, worsen infant botulism.[116] Lysis of *C. botulinum* in the gut by antibiotics may also increase the toxin available in infant botulism.[117] This effect has not been reported in adult cases but should be considered when gastrointestinal infection is suspected. Aminopyridines promote neurotransmission from intoxicated nerve terminals and may have a role in the treatment of the paralysis associated with botulism.[118]

In the event of a bioterrorist attack, several logistical issues would make treatment problematic. Based on the limited information available, a large-scale attack (either foodborne or by aerosol) would probably not begin to produce symptomatic victims for more than a day, thereby delaying diagnosis and containment. Furthermore, the supply of antitoxins is small, reflecting the low incidence of the natural disease. With a large-scale attack, the available antitoxin would be quickly exhausted. Thus, airway protection and ventilation may be the only viable treatment options. To minimize additional exposures, exposed skin and clothing should be washed with soap and water, whereas contaminated surfaces should be cleaned with 0.1% hypochlorite bleach solution if they cannot be avoided for the hours to days required for natural degradation.[48]

Although the greatest improvement in muscle strength occurs in the first 3 months of recovery from botulism, patients still show improvements in strength and endurance for up to 1 year after disease onset.[48,119] With prompt attention and supportive care, the mortality rate for botulism ranges from less than 5% to 8%.[119] The mortality rate for infant botulism is less than 1%.[28]

Long-term consequences of botulism were detailed in an evaluation of 211 patients from the Republic of Georgia from 1998 to 2003. Patients interviewed at least 6 months after illness reported higher rates of fatigue, weakness, and dyspnea on exertion when compared with controls. Affected patients had limitations in functional capacity and impaired psychosocial well-being.[120]

PREVENTION

The most important aspect of botulism prevention is proper food handling and preparation. It is impractical or undesirable to treat many foods in a manner to eliminate *C. botulinum* spores; hence, methods for the control of botulism focus on the inhibition of bacterial growth and toxin production.[117] Because the toxin is heat labile, terminal boiling or similarly intense heating of contaminated food will inactivate it. Food containers that appear to bulge may contain gas produced by *C. botulinum* and should not be opened. Other foods that appear to be spoiled should not be tasted.

In the event of an outbreak, foods suspected of being contaminated should be refrigerated until retrieval by public health personnel. Laboratory testing for botulism in the United States is available only at the CDC and several state and city public health laboratories. According to the Working Group on Civilian Biodefense, persons with potential exposure in a foodborne botulism outbreak should be monitored closely for the development of signs and symptoms; antitoxin should be administered promptly at the first signs of illness.[48]

Immunity to botulinum toxin does not develop even with severe disease, and the repeated occurrence of botulism has been reported.[121] An investigational pentavalent toxoid (ABCDE) vaccine was previously available for use among high-risk laboratory workers and military personnel in the United States.[122] However, in 2011 the CDC discontinued use of the vaccine because of data suggesting waning immunogenicity and frequent local reactions.[123] New vaccine strategies, including recombinant toxin A/B vaccines and inhalational vaccines, are currently being investigated.[54,124]

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246 Diseases Caused by *Clostridium*

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SHORT VIEW SUMMARY

CHARACTERISTICS OF *CLOSTRIDIUM* SPECIES

- Member of the phylum Firmicutes
- Anaerobic gram-positive rods capable of forming endospores
- Ubiquitous in nature
- Traditional phenotypic classification methods rely on carbohydrate fermentation, detection of short-chain fatty acid end products of fermentation, Gram stain morphology, colony morphology, and detection of specific toxins.

CLOSTRIDIOIDES (FORMERLY CLOSTRIDIUM) DIFFICILE INFECTION (CDI)

- Clinical manifestations range from a self-limiting diarrheal disease that disappears when antibiotics are discontinued to fulminant presentations with characteristic pseudomembranes within the large intestine and progression to toxic megacolon and fatal complications.
- The initiating event involves the disruption of the intestinal microbiome during antibiotic treatment, followed by the germination of *C. difficile* spores and production of an enterotoxin (TcdA) and a cytotoxin (TcdB).
- Highly virulent toxin-producing strains possess a deletion of TcdC, which normally downregulates toxin production.
- Treatment includes metronidazole in less serious cases, vancomycin for serious and/or progressive disease, and fecal transplantation for recurrent disease.
- Diagnosis: enzyme immunoassays for the detection of TcdA and/or TcdB, membrane immunoassay to detect the antigen glutamate dehydrogenase and toxin, and polymerase chain reaction.
- Infection control measures, including rapid diagnosis of CDI, the use of contact precautions for positive patients, appropriate therapy, and rigorous cleaning of rooms to effectively kill spores, must be implemented with the hospital or nursing home environment.

CLOSTRIDIUM PERFRINGENS AND CLOSTRIDIAL MYONECROSIS (GAS GANGRENE)

- *Clostridium perfringens* produces a variety of toxins (see Table 246.2). All strains produce α-toxin, a lecithinase that causes cell membrane damage.
- Most common following traumatic crushing injures that result in lowered tissue oxygen levels, penetrating trauma involving foreign bodies contaminated with soil, gastrointestinal or biliary tract surgery, and septic abortion.
- Diagnosis: Initial symptoms include severe pain, redness at the wound site followed by rapidly spreading brown to purple discoloration, edema and gas, and serosanguineous discharge with a characteristic "mousy" odor. Progression to full-blown sepsis with hypotension, renal failure, and metabolic acidosis occurs rapidly.
- Treatment involves prompt surgical débridement of infected tissues, including amputation for extremities or hysterectomy in uterine gas gangrene. Antibiotic treatment with penicillin, metronidazole, clindamycin, or the carbapenems.

FOOD POISONING CAUSED BY *CLOSTRIDIUM PERFRINGENS*

- *Clostridium perfringens* type A is involved in most cases.
- Involves the ingestion of at least 10^8 viable enterotoxin-producing cells in food products that are not properly cooked or stored.
- The incubation period is 7 to 15 hours, and cases resolve spontaneously within 24 to 48 hours.

OTHER CLOSTRIDIAL INFECTIONS

- Bacteremia—Clostridia account for 1% of all positive blood cultures. Significant risk factors include hemodialysis, intestinal malignancy, and inflammatory bowel disease.
- Biliary tract infections—Clostridia can be isolated from over 20% of diseased gallbladders, and *C. perfringens* accounts for 50% of these. Radiographic detection of gas within the biliary tract requires surgical intervention and prompt antibiotic treatment.
- Female genital tract infections—Clostridia are present in up to 20% of non–sexually transmitted disease genital infections and may be present as constituents of bacterial vaginosis. *Clostridium perfringens* and *Clostridium sordellii* can be isolated from postpartum and postabortion infections.
- Pleuropulmonary infections—Clostridia are recovered from up to 10% of pulmonary infections, with *C. perfringens* accounting for the majority.

The genus *Clostridium* includes over 200 described species. Members of this genus participate in a variety of invasive and toxigenic infections. They can cause disease that is strictly toxin mediated, such as antibiotic-associated colitis (AAC) and foodborne botulism, or contribute to invasive infections, including bacteremia, clostridial myonecrosis (gas gangrene), and other suppurative infections driven by histotoxins and enzymes that destroy soft tissue. Historically, clostridial infections were recognized as discrete clinical syndromes well before the germ theory of disease was proposed. The clinical features of tetanus were well described by some of the earliest medical writers, such as Hippocrates, and the toxic nature of this species was noted as early as the 1870s.[1] Clostridia are often isolated as part of a mixed microbiota during suppurative infections that occur as a result of fecal or soil contamination of otherwise sterile tissues. Prior to 1977, the most commonly reported clostridial infections and intoxications were those caused by *Clostridium perfringens*. Other species, most notably *Clostridium tetani* in nonimmunized individuals and *Clostridium botulinum*, also generated considerable interest due to the severity and often fatal nature of the intoxication they caused.

With the discovery of the etiology of AAC first in an animal model[2] and subsequently in humans,[3] it soon became clear that in the antibiotic era *Clostridioides difficile (formerly Clostridium difficile)* was the most common clostridial species associated with the human disease formerly called C. *difficile*–associated diarrhea and currently called *C. difficile* infection (CDI). Within the hospital setting, CDI has become a significant worldwide nosocomial infection problem[4] resulting in both toxin-mediated diarrheal disease and more fulminant presentations such as pseudomembranous enterocolitis and toxic megacolon. Recent recognition that more virulent strains of *C. difficile* occur in health care settings has provoked an increased awareness of this nosocomial infection and has prompted a demand for both rapid methods for diagnosis and more aggressive treatment of CDI and AAC.[5–7] While the well-recognized

pathogenic members of the genus *Clostridium* continue to participate in a broad array of infectious processes, it is also important to note the important role that previously obscure species, such as *C. difficile,* play in human disease.

CHARACTERISTICS OF *CLOSTRIDIUM* SPECIES

Microbiology

Members of this genus are phenotypically characterized as anaerobic, gram-positive rods capable of forming endospores. *Clostridium* spp. are ubiquitous in nature, found in soils and sediments throughout the world and as members of the intestinal microbiome of humans and most other animals. Over 70% of humans are colonized with clostridia at concentrations of 10^8 to10^9 organisms per gram of feces.[8] Clostridia can also be isolated as part of the vaginal microbiome of healthy women,[9] although they tend to be transient members of this microbiome, occurring in low numbers as a result of contamination by intestinal microbiota, rather than as part of the autochthonous community. Most members of this genus are obligate anaerobes, while strains of a few *Clostridium* species, such as *C. tertium, C. histolyticum, C. innocuum,* and *C. perfringens,* are aerotolerant and can be confused with members of the genus *Bacillus* during laboratory diagnosis. Based on 16S ribosomal DNA (rDNA) sequence data, members of the genus *Clostridium* are part of the phylum Firmicutes, a diverse group of gram-positive organisms including both spore-forming and non–spore-forming genera. Based on 16S rDNA sequence analysis, the clostridia can be divided into 11 homology groups, with most of the clinically significant species belonging to homology group 1.[10] Traditional phenotypic classification methods for the clostridia rely on carbohydrate fermentation profiles, detection of short-chain fatty acid end products of fermentation, Gram stain morphology, colony morphology on agar media, and detection of specific toxins. More recently, proteomic analysis using matrix-assisted laser desorption/ionization time-of-flight (MALDI-TOF) mass spectrometry has been employed for identification of certain species.[11,12] Although many different species have been isolated from human clinical material, only a small number of species are regularly associated with human disease (Table 246.1).

Microscopically, the vegetative cells of *Clostridium* species are rod-shaped, often pleomorphic, and found as short chains, as clusters, or in pairs. The cells of most species have rounded ends. This may vary, with some species showing more pointed ends (*Clostridium ramosum*). Some species form long chains (*Clostridium spiroforme*), which may be tightly packed to form coils. Clostridia usually stain gram positively in young cultures, with some species losing this staining characteristic in older cultures. Species such as *Clostridium clostridioforme* and *C. ramosum* rarely show the typical gram-positive appearance and present as gram-variable or gram-negative rods (Fig. 246.1). When spores are present, they tend to be ovoid or spherical, with the spore often distending the vegetative cell to produce a "club-shaped" appearance. Spores may be located centrally, subterminally, or as terminal structures, depending on the species. Spore location is used as part of the phenotypic identification process. Most clostridia are motile by virtue of peritrichous flagellae, with the notable exception of the common clinical isolates *C. perfringens* and *C. ramosum*.[10]

Clostridium spp. have diverse metabolic pathways and can be saccharolytic, proteolytic, both, or neither. *Clostridium* spp. are not known to reduce sulfate. The end products of fermentative metabolism are mixtures of short-chain fatty acids and alcohols, a characteristic that can be used for identification purposes in the clinical laboratory. Aerotolerant strains of clostridia do not form spores in the presence of oxygen, are catalase negative, and grow more abundantly under anaerobic conditions. Clostridia do not have a complete cytochrome system and are therefore oxidase negative.[10] Most strains are catalase and superoxide dismutase negative, although trace amounts of activity have been reported for some species. Clostridia produce a variety of biologically active proteins, including hemolysins, proteolytic enzymes, and other toxins. It is the protein toxins produced by clostridia that account for their importance in human disease. Clostridia produce a greater diversity of toxins than any other genera of bacteria.[10] These include neurotoxins, enterotoxins, collagenases, proteases, necrotoxins, lecithinases, lipases, DNases, and neuraminidases. The potency of some of these toxins, such as botulinum neurotoxin (BoNT) and tetanus neurotoxin (TeNT), render them among the most lethal substances yet described; less than 0.2 ng of purified TeNT is fatal in mice.

Pathogenesis

Invasive infections caused by clostridia are invariably due to organisms that are either part of the normal intestinal and vaginal microbiome or acquired by a traumatic injury breaching the skin that becomes contaminated with soil, unsanitary water, or fecal material. Intoxications can occur either in response to endogenous toxin production, such as that associated with CDI, or by ingestion of preformed toxins

TABLE 246.1 Clostridial Species Commonly Associated With Human Disease

SPECIES	SPORE LOCATION	LECITHINASE PRODUCED	LIPASE	ENTEROTOXINS PRODUCED	HISTOTOXINS, HEMOLYSINS, PROTEASES	NEUROTOXINS PRODUCED
Tissue Infections						
C. perfringens	ST, C	+	–	Yes	Yes	No
C. ramosum	T	–	–	No	Yes	No
C. septicum	ST	–	–	No	Yes	No
C. sordellii	ST	+	–	No	Yes	No
C. bifermentans	ST	+	–	No	Yes	No
C. tertium	T	–	–	No	Yes	No
C. sphenoides	ST	–	–	No	Yes	No
C. baratii	ST	–	–	No	Yes	No
C. novyi	ST	+	+	No	Yes	No
C. histolyticum	ST	–	–	No	Yes	No
Intoxications						
C. difficile	ST	–	–	Yes	Yes	No
C. botulinum	ST, T	–	+	No	Yes	Yes
C. tetani	T	–	–	No	Yes	Yes

C, Centrally; *ST,* subterminally; *T,* terminally.

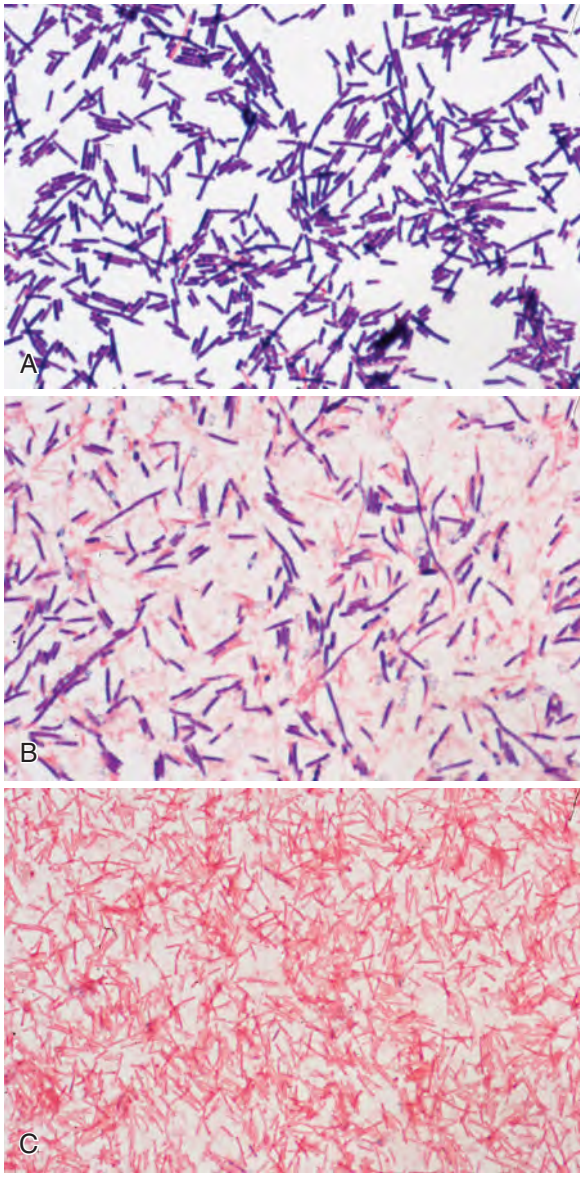

FIG. 246.1 Gram stain characteristics of *Clostridium* species. (A) *Clostridium perfringens.* (B) *Clostridium novyi.* (C) *Clostridium ramosum.*

contaminating food, as is the case for noninfant botulism. Apart from environmental spread of *C. difficile* within a susceptible population, such as hospitalized patients on broad-spectrum antibiotic therapy and residents in nursing homes, clostridia rarely cause infection through person-to-person contact.

The spores of clostridia account for their persistence in hostile environments and their exogenous acquisition by humans. In addition to their long-term survival in soil or food, clostridial spores may spread via aerosol transmission as part of naturally occurring dust clouds. *C. difficile* is of concern because this species may be part of the intestinal microbiome of an individual or may be acquired through contact with individuals or contaminated surfaces and equipment within the hospital environment harboring spores. The vegetative cells of clostridia are generally susceptible to routinely used disinfectants; however, spores can survive hostile environments, including heat, desiccation, and exposure to many commonly employed disinfectants.[13–15] This allows pathogenic clostridia to persist in the environment, even following routine disinfection procedures. Methods for eliminating clostridial spores from some environments, such as *C. difficile* in the hospital setting, include finding methods to promote germination of the spores so that the vegetative cells can be destroyed.[13,16]

Clostridium spp., particularly members of clusters IV and XIVa derived from the gut microbiota, have recently been shown to have beneficial effects on the immune system. One can speculate that this may be why almost all children have *C. difficile* present in their intestine during the first year of life. Specifically, a consortia of *Clostridium* species promoted accumulation of colonic regulatory T-cell development in mice.[17,18] Inoculation of these *Clostridium* spp. into mice increased their resistance to chemically induced colitis and blunted their allergic responses. This finding casts this genus in a new light, suggesting its members are not only fearsome pathogens but also may hold therapeutic potential for autoimmune and allergic conditions.

MAJOR INFECTIONS AND INTOXICATIONS

Clostridioides difficile Infection

Historical Perspective

Until it was identified as the primary cause of AAC in 1977, *C. difficile* was not regarded as a particularly common or important pathogen; however, the association of *C. difficile* with CDI and AAC has brought this organism to prominence as the most common clostridial species associated with human disease. Hall and O'Toole published the first description of *C. difficile* in 1935 and suggested that it might be involved in intestinal disease in children.[19] Interestingly, the clinical description of pseudomembranous colitis dates to the 1890s. An animal model for antibiotic-associated intestinal disease was first reported in the 1940s, with several additional observations on the induction of bowel inflammation by antibiotics in hamsters, guinea pigs, and rabbits made in the 1950s. The occurrence of pseudomembranous colitis in patients receiving broad-spectrum antibiotics prior to the 1970s was not uncommon. Based on laboratory analysis, it was often attributed to *Staphylococcus aureus,* one of the major nosocomial pathogens of the antibiotic era. Cultures of stool from these patients often yielded high levels of *S. aureus;* however, obligately anaerobic organisms were not evaluated in these early studies. Given the common isolation of *S. aureus* from stool samples obtained from healthy individuals, these earlier observations were something of a self-fulfilling prophecy. While certain strains of *S. aureus* produce potent enterotoxins that may be responsible for some cases of AAC, there is little evidence to suggest that this species is a common cause of pseudomembranous colitis.

In 1974, investigators in St. Louis noted that about 20% of patients receiving the lincosamide antibiotic clindamycin developed diarrhea, and half of these patients had pseudomembranous colitis when examined endoscopically.[20] Publication of these observations set the stage for more detailed examination of the role of antibiotics in the occurrence of pseudomembranous colitis and led to the search for an etiologic agent. The breakthrough that ultimately led to identification of *C. difficile* as the causative agent of CDI involved a hamster model of AAC demonstrating that vancomycin prevented the occurrence of AAC induced by clindamycin, suggesting that a gram-positive organism was involved in the hamster disease.[21] Armed with this information and the knowledge that the disease appeared to be toxin-mediated, based on molecular size filtration studies, these same investigators isolated clostridial species from the ceca of hamsters with AAC and showed that one species, *C. difficile,* can cause disease in other hamsters as pure cultures or culture filtrates in the absence of prior antibiotic exposure. The link to human disease was made when the same toxin isolated from the hamster model was found in stools of AAC patients using a combination of cytotoxicity assays and an anti-clostridial antibody capable of neutralizing the cytotoxic effect.[22] Vancomycin remains the antibiotic of choice for treating serious CDI in humans (see Chapter 243 for a more extensive discussion of treatment alternatives), and the same cytotoxicity assay used to correlate animal and human disease remains the gold standard for evaluating other diagnostic assays.

Clinical Manifestations

The clinical manifestations of CDI range from a self-limiting diarrheal disease that disappears when antibiotics are discontinued to fulminant presentations with characteristic pseudomembranes within the large intestine and progression to toxic megacolon, often with fatal complications.[23] Pseudomembranes, while present in 97% of CDI, are not