Chapter 25

EMERGING INFECTIOUS DISEASES AND FUTURE THREATS

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INTRODUCTION

Emerging infectious diseases, as defined in the landmark 1992 report by the Institute of Medicine, are diseases whose incidence has increased within the past 20 years or whose incidence threatens to increase in the near future.[1] Even though some "emerging" diseases have now been recognized for over 20 years (eg, acquired immunodeficiency syndrome [AIDS], Lyme disease, Legionnaire's disease), their importance has not diminished, and the factors associated with their emergence are still relevant. Emerging infections include diseases caused by new agents (or newly described agents) and reemerging pathogens (those whose incidence had previously declined but now is increasing). This definition also includes organisms that are developing antimicrobial resistance and established diseases with a recently discovered infectious origin.

Many factors contribute to the emergence of new diseases. In the United States, in particular, these factors include increasing population density and urbanization; immunosuppression (resulting from aging, malnutrition, cancer, or infections such as AIDS); changes in land use (eg, deforestation and reforestation), climate, and weather; international travel and commerce; and microbial or vector adaptation and change (mutations which result in drug or pesticide resistance).[1] Internationally, many of these factors also hold true; however, many developing countries also have to deal with war, political instability, inadequate healthcare, and basic sanitation issues.

The numerous examples of "new" infections originating from animal species (ie, zoonoses) suggest that the zoonotic pool is an important and potentially rich source of emerging diseases.[2] Although classifying AIDS as a zoonotic disease is controversial,[3] it is now clear that both human immunodeficiency virus [HIV]-1 and HIV-2 had zoonotic origins.[4-6] In addition, as shown by the 2003 outbreak of monkeypox in the United States, increasing trade in exotic animals for pets has led to increased opportunities for pathogens to "jump" from animal reservoirs to humans. The use of exotic animals (eg, Himalayan palm civets) for food in China and the close aggregation of numerous animal species in public markets may have led to the emergence of the severe acute respiratory syndrome (SARS) coronavirus.[7]

Many of the viruses or bacteria that may be potential bioweapons are considered emerging pathogens. In particular, some of these agents have appeared in new geographical locations where they have not previously been seen; for example, monkeypox suddenly occurred in the US Midwest in 2003, and the largest recorded outbreak of Marburg hemorrhagic fever occurred in Angola in 2005. Sometimes the specific use of a pathogen in an act of bioterrorism can cause the pathogen to be classified as an emerging or reemerging disease agent, as what happened with *Bacillus anthracis* during the 2001 anthrax attacks in the United States. Through increasingly easy molecular biology techniques, completely new organisms (or significantly modified existing organisms) can now be made in the laboratory. The use of these methods is mostly beneficial and necessary for modern biomedical research to proceed. However, the same methods and techniques can be used for destructive purposes and, along with naturally occurring emerging infections, represent significant future threats to both military and civilian populations.

EMERGING BACTERIAL DISEASES

Waterborne Diseases

Emerging waterborne diseases constitute a major health hazard in both developing and developed countries. In 2001 and 2002, 31 disease outbreaks associated with contaminated drinking water were reported in the United States, resulting in 1,020 ill people and 7 deaths.[8] During this same time, over 2,500 cases of illness and 8 deaths nationally were associated with recreational waterborne diseases.[9] Bacterial pathogens associated with drinking water disease outbreaks included *Legionella* species, *Escherichia coli* O157:H7, and *Campylobacter jejuni* (one outbreak each), and one outbreak involving infection with two different bacteria: *C jejuni* and *Yersinia enterolitica*.[8] Bacterial pathogens responsible for gastroenteritis outbreaks associated with recreational water exposure included *E coli* O157:H7 (four outbreaks) and *Shigella sonnei* (two outbreaks). Twenty dermatitis outbreaks associated with spa or pool use were attributed to *Pseudomonas*, primarily *P aeruginosa*.[9]

Vibrio cholerae and Cholera

Accounts of cholera date to Hippocrates.[10] Seven worldwide cholera pandemics have occurred. An 1892 cholera outbreak in Hamburg, Germany, affecting 17,000 people and causing 8,605 deaths was attributed to the inadvertent contamination of the city's water supply by bacteriologists studying the pathogen.[11] This

event underscores the potential for cholera to cause widespread illness where water is not disinfected with a modern bacteriocide such as chlorine.[11]

In 1991, after almost a century without cholera, outbreaks in Latin America resulted in about 400,000 cases of cholera and over 4,000 deaths.[12] Off the Peruvian coast, a significant correlation between cholera incidence and elevated sea surface temperature occurred between 1997 and 2000, which included the 1997–1998 El Niño event.[13] Some people believe that the eighth worldwide pandemic began in 1992 with the emergence and spread of a new epidemic-causing strain (see below).[14] During 2003, 45 countries reported a total of about 112,000 cases and almost 1,900 deaths from cholera.[15] Paradoxically, cholera cases in the United States have decreased to about 10 cases per year during 1995 through 2000. Most of these cases were associated with either travel or consumption of undercooked seafood harvested along the Gulf coast.

Cholera occurs through fecal-oral transmission brought about by deterioration of sanitary conditions. Epidemics are strongly linked to the consumption of unsafe water, poor hygiene, poor sanitation, and crowded living conditions (Figure 25-1). Water or food contaminated by human waste is the major vehicle for disease transmission. Cholera transmission is thought to require 10^3 organisms to exert an effect in the gut, with 10^{11} organisms as the minimum infective dose able to survive stomach acid.[16]

Before 1992, all cholera pandemics were caused by the *V cholerae* serogroup O1 (classical) or El Tor biotypes. Large outbreaks in 1992 resulted from transmission of a previously unknown serogroup, *V cholerae* O139, which has since spread from India and Bangladesh to countries throughout Asia, including Pakistan, Nepal, China, Thailand, Kazakhstan, Afghanistan, and Malaysia.[17,18] Cholera vaccines have had mixed success. Historically, live attenuated vaccines are more effective than killed whole-cell vaccines.[19] No licensed cholera vaccines are available in the United States.

Enterotoxin produced by *V cholera* O1 and O139 can cause severe fluid loss from the gut. In severe cases, profuse watery diarrhea, nausea, and vomiting can lead to rapid dehydration, acidosis, circulatory collapse, and renal failure. Successful treatment of cholera patients depends on rapid fluid and electrolyte replacement; antimicrobial therapy can also be useful.

Other Vibrios

In recent years, some noncholera vibrios have acquired increasing importance because of their association with human disease. Over 70 members are in the family *Vibrionaceae*, 12 of which have been isolated from human clinical specimens and apparently are pathogenic for humans.[20] *Vibrio* species are primarily aquatic and are very common in marine and estuarine environments and on the surface and in the intestinal tracts of marine animals. *V parahaemolyticus* and *V vulnificus* are halophilic vibrios commonly associated with consumption of undercooked seafood. Diarrhea, cramping, nausea, vomiting, fever, and headache are commonly associated with *V parahaemolyticus* infections. *V vulnificus*, the most common source of vibrio infections in the United States, results in gastrointestinal symptoms similar to those of *V parahaemolyticus* and may also lead to ulcerative skin infections if open wounds are exposed to contaminated water. Septicemia can also occur in infected persons who are immunosuppressed or have liver disease or chronic alcoholism, and septicemic patients can have a mortality rate of up to 50%. In most cases the disease begins several days after the patient has eaten raw oysters. Other human pathogenic species include *V mimicus*, *V*

Fig. 25-1. Typical conditions that can lead to a cholera epidemic. This photograph was taken in 1974 during a cholera research and nutrition survey amidst floodwaters in Bangladesh.
Photograph: Courtesy of Dr Jack Weissman, Centers for Disease Control and Prevention Public Health Image Library.

metschnikovii, *V cincinnatiensis*, *V hollisae*, *V damsela*, *V fluvialis*, *V furnissii*, *V alginolyticus*, and *V harveyi;* most of these have been associated with sporadic diarrhea, septicemia, and wound infections.[20]

Legionella Species

Legionnaire's disease was first recognized in 1976 after a large outbreak of severe pneumonia occurred among attendees at a convention of war veterans in Philadelphia. A total of 182 people, all members of the Pennsylvania American Legion, developed an acute respiratory illness, and 29 individuals died from the disease.[21] The cause of the outbreak remained a mystery for 6 months until the discovery by Joseph McDade, a Centers for Disease Control and Prevention microbiologist, of a few gram-negative bacilli, subsequently named *Legionella pneumophila*,[22] in a gram stain of tissue from a guinea pig inoculated with lung tissue of a patient who died from the disease.[23] Using the indirect immunofluorescence assay, McDade showed that the sera of patients from the convention mounted an antibody response against the newly isolated bacterium,[24] marking the discovery of a whole new family of pathogenic bacteria. Retrospective analysis, however, has shown that outbreaks of acute respiratory disease from as far back as 1957 can be attributed to *L pneumophila*.[24,25] The earliest recorded isolate of a *Legionella* species was recovered by Hugh Tatlock in 1943 during an outbreak of Fort Bragg fever.[26,27]

Legionnaire's disease is normally acquired by inhalation or aspiration of *L pneumophila* or other closely related *Legionella* species. Water is the major reservoir for legionellae, and the bacteria are found in freshwater environments worldwide. Legionnaire's disease has been associated with various water sources where bacterial growth is permitted, including cooling towers,[28] whirlpool spas,[29] and grocery store mist machines.[29] The association between a potable shower and nosocomial legionellosis was demonstrated 25 years ago.[30] The most common source of legionellosis in hospitals is from the hot water system,[31] and sustained transmission of Legionnaire's disease in the hospital can be difficult to control.[32] Community-acquired legionellosis is thought to account for most infections.[33] A recent Italian survey of household hot water systems found bacterial contamination with *Legionella* species in 23% of the homes and *Pseudomonas* species in 38%. One *Legionella* species, *L longbeachae*, has been associated with disease transmission from potting soil.[34]

Legionnaire's disease is an acute bacterial illness. Patients initially present with anorexia, malaise, myalgia, and headache, with a rapidly rising fever and chills. Temperatures commonly reach 102°F to 105°F and are associated with nonproductive cough, abdominal pain, and diarrhea. The disease may eventually progress to respiratory failure and has a case-fatality rate as high as 39% in hospitalized cases. Nonpneumonic legionellosis, or Pontiac fever, occurs after exposure to aerosols of water colonized with *Legionella* species.[35-37] Attack rates after exposure to an aerosol-generating source are exceptionally high, often in the range of 50% to 80%. After a typical asymptomatic interval of 12 to 48 hours after exposure, patients note the abrupt onset of fever, chills, headache, malaise, and myalgias. Pneumonia is absent, and those who are affected recover in 2 to 7 days without receiving specific treatment.[38]

Legionella is now recognized around the world as an important cause of community-acquired and hospital-acquired pneumonia, occurring both sporadically and in outbreaks. Although 90% of *Legionella* infections in humans are caused by *L pneumophila*, there are 48 named species of *Legionella*, with at least 20 known to cause human infections.[39] Some unusual strains of bacteria, which infect amoebae and have been termed *Legionella*-like amoebal pathogens (LLAPs), appear to be closely related to *Legionella* species on the basis of 16S ribosomal RNA gene sequencing.[40,41] Three LLAP strains are now named *Legionella* species,[42] and one of them, LLAP-3, was first isolated from the sputum of a patient with pneumonia by coculture with amoebae and is considered a human pathogen.[43]

Foodborne Diseases

More than 200 diseases are transmitted through food, including illnesses resulting from viruses, bacteria, parasites, toxins, metals, and prions. In the United States the burden of foodborne illness is estimated at approximately 76 million illnesses, 325,000 hospitalizations, and 5,000 deaths each year.[44] Among the bacterial pathogens estimated to cause the greatest number of US foodborne illnesses are *Campylobacter*, *Salmonella*, *Shigella*, *Clostridium*, and *Staphylococcus* species.[44] Emerging bacterial illnesses include *E coli* O157:H7 and other enterohemorrhagic and enterotoxigenic *E coli*, as well as antibiotic-resistant bacteria. Many of the pathogens of greatest concern today (eg, *C jejuni*, *E coli* O157:H7, *Listeria monocytogenes*, *Cyclospora cayetanensis*) were not recognized as causes of foodborne illness just 20 years ago, and some proportion of gastrointestinal illness is caused by foodborne agents that have not yet been identified. It is estimated that 62 million foodborne-related illnesses and 3,200 deaths occur in the United States each year from unknown pathogens.[44] *Bacillus anthracis*, although rarely seen as a gastrointestinal illness in the United States, has become a concern since cases occurred in 2000 (see

below). Even in areas of the world where gastrointestinal anthrax is more common, the oropharyngeal form is underreported because physicians are unfamiliar with it.[45] Unreported foodborne disease, deaths from unknown food agents,[46] and chronic sequelae[47] may be a huge unrecognized burden of illness.

Bacillus anthracis

B anthracis is the causative agent of anthrax, a naturally occurring zoonotic disease. The greatest bioweapon threat from anthrax is through aerosol dispersion and subsequent inhalation of concentrated spores (for more details see Chapter 4, Anthrax). Gastrointestinal anthrax, however, is contracted through the ingestion of *B anthracis* spores in contaminated food or water. This form of the disease occurs more commonly than inhalational anthrax in the developing world, but is rare in the United States and other developed nations.[45,48] In one large outbreak in Uganda, 155 villagers ate the meat of a zebu (bovine) that had died of an unknown disease. Within 15 to 72 hours, 143 persons (92%) developed presumed anthrax. Of these, 91% had gastrointestinal complaints and 9% had oropharyngeal edema; 9 of the victims, all children, died within 48 hours of illness onset.[48] Gastrointestinal anthrax can occur naturally in the United States, and anthrax-contaminated meat has been found to be associated with gastrointestinal illness in Minnesota as recently as 2000.[49] Purposeful contamination of food or water is possible but would require a high infective dose. Misdiagnosis may lead to a higher mortality in gastrointestinal anthrax than in other forms of the disease; thus, awareness of this disease remains important in anthrax-endemic areas and in the setting of possible bioterrorism.

Campylobacter jejuni

Campylobacter was first identified in 1909 (then called *Vibrio fetus*) from the placentas and aborted fetuses of cattle. The organism was not isolated from humans until nearly 40 years later, when it was found in the blood of a pregnant woman who had an infectious abortion in 1947.[50] *Campylobacter jejuni* (Figure 25-2), along with *C coli*, have been recognized as agents of gastrointestinal infection since the late 1970s. *C jejuni* is considered the most commonly reported foodborne bacterial pathogen in the United States, affecting 2.4 million persons annually.[51] Campylobacteriosis is an enteric illness of variable severity, including symptoms of diarrhea (which may be bloody), abdominal pain, malaise, fever, nausea, and vomiting, occurring 2 to 5 days after exposure. Although many infections are asymptomatic, infection with this pathogen has been associated with development of Guillain-Barré syndrome and arthritis.[52,53] Infants are more susceptible to *C jejuni* infections upon first exposure.[54] Persons who recover from *C jejuni* infection develop immunity. Poultry colonized with *Campylobacter* species is a major source of infections for humans.[55-58] The reported incidence of *Campylobacter* species on poultry carcasses has varied but has been as high as 100%.[57]

Several virulence properties, including motility, adherence, invasion, and toxin production, have been recognized in *C jejuni*.[59] Along with several other enteric bacteria, *C jejuni* produces a toxin called cytolethal distending toxin that works by a completely novel mechanism: mammalian cells exposed to the toxin distend to almost 10 times their normal size from a molecular blockage in their cell cycle.[60] Although cytolethal distending toxin is the best-characterized *Campylobacter* toxin, its role in the pathogenesis of human campylobacteriosis is unclear.[61]

Because illness from *Campylobacter* infection is generally self limited, no treatment other than rehydration and electrolyte replacement is generally recommended. However, in more severe cases (ie, with high fever, bloody diarrhea, or septicemia), antibiotic therapy can be used to shorten the duration of symptoms if it is given early in the illness. Because infection with *C jejuni* in pregnant women may have deleterious effects on the fetus, infected pregnant women should receive antimicrobial treatment. Erythromycin, because it is safe, lacks serious toxicity, and is easy to administer, is the drug of choice for *C jejuni* infections. However, most clinical trials performed in adults or children

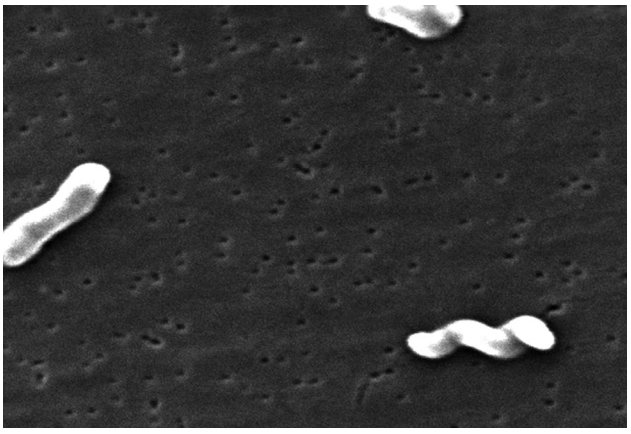

Fig. 25-2. Scanning electron microscope image of *Campylobacter jejuni* illustrating its corkscrew appearance.
Photograph: Courtesy of Janice Carr, Centers for Disease Control and Prevention Public Health Image Library.

have not found that erythromycin significantly alters the clinical course of *Campylobacter* infections.[62,63] Other antimicrobial agents, particularly the quinolones (eg, fluoroquinolones such as ciprofloxacin) and newer macrolides including azithromycin are also being used. Unfortunately, as the use of fluoroquinolones has expanded (especially in food animals), the rate of resistance of campylobacters to these agents has increased.[64] For example, a 1994 study found that most clinical isolates of *C jejuni* from US troops in Thailand were resistant to ciprofloxacin. Additionally, nearly one third of isolates from US troops located in Hat Yai were resistant to azithromycin.[65] In another study conducted in 1997 in Minnesota, 13 of 91 chicken products (14%) purchased in grocery stores were contaminated with ciprofloxacin-resistant *C jejuni*,[66] illustrating the need for more prudent antimicrobial use in food-animal production.

Clostridium botulinum

C botulinum produces botulinum toxin, which causes the clinical manifestations of botulism. Botulinum toxin, with a lethal dose of about 1 μg/kg, is the most potent of the natural toxins.[67] There are seven antigenic types of toxin, designated A through G; most human disease is caused by types A, B, and E. Botulinum toxins A and B are most often associated with home canning and home-prepared foods, while botulinum toxin E is exclusively associated with ingestion of aquatic animals. The incidence of botulism in Alaska is among the highest in the world, and all cases of foodborne botulism in Alaska have been associated with eating traditional Alaska Native foods, mostly from marine mammals; most of these cases were caused by toxin type E.[68] From 1990 to 2000, 160 foodborne botulism events affected 263 persons in the United States. Of these patients, 67 required intubation, and 11 deaths occurred.[69] Food items commonly associated with botulinum intoxication included homemade salsa and home-bottled garlic in oil.

Clinical illness is characterized by cranial nerve palsies, followed by symmetric descending flaccid muscle paralysis, which may involve the respiratory muscles. Full recovery may take weeks to months. Therapy includes intensive care support, mechanical ventilation as necessary, and timely administration of equine antitoxin.[69]

Escherichia coli O157:H7

E coli O157:H7 has emerged as a cause of serious pediatric illness worldwide. Its intrinsic Shiga toxins can initiate a cascade of events that include bloody diarrhea and hemolytic uremic syndrome (HUS), exhibited by microangiopathic hemolytic anemia, acute renal failure, and thrombocytopenia.[70] HUS occurs in about 4% of all reported cases, and persons under five years of age are at greatest risk.[44] The mortality rate for HUS is 3% to 5%, and about 5% of survivors have severe consequences, including end-stage renal disease and permanent neurological damage.[71] Antibiotic treatment of *E coli* O157:H7 is not recommended.[72] There is anecdotal evidence for an increase in the risk of HUS with the use of some antimicrobial agents, although conclusive proof of this occurrence is lacking. Fluid replacement is the cornerstone of the treatment of diarrheal illness caused by the enterohemorrhagic *E coli*.

The primary source of *E coli* O157:H7 is beef cattle. The current animal culture practice of feeding grain (rather than hay) to these animals decreases the pH in their colons, thereby promoting acid-resistance and enhanced growth of *E coli* pathogens.[73]

Salmonella Species

Salmonella species infect an estimated 1.4 million persons annually in the United States. Although most infections are self-limiting, with diarrhea, vomiting, abdominal cramps, and fever, severe infections are not uncommon. Estimates suggest that approximately 15,000 people are hospitalized and over 500 deaths occur each year from *Salmonella* infections.[44] Food animals are the primary reservoir for human nontyphoidal *Salmonella* infections. There are thousands of *Salmonella* serotypes, and many naturally inhabit avian, mammalian, and reptilian gastrointestinal tracts. Poultry is the main source of the salmonellae in the food supply; other vehicles for disease transmission include raw salads, milk, water, and shellfish.

Infection with many *Salmonella* serotypes causes gastroenteritis with associated diarrhea, vomiting, febrile illness, headache, and dehydration. Septicemia, enteric fever, and localized infections may also evolve from salmonellosis infection. The most highly pathogenic of the salmonellae, *S typhi*, causes typhoid fever, which includes symptoms of septicemia, high fever, headache, and gastrointestinal illness. *S typhimurium* was the pathogen used in 1984 by an Oregon cult to cause illness by purposeful contamination of salad bars.[74] Over 750 cases of illness resulted, but no deaths occurred, which may not have been the case if *S typhi* had been used. A 1985 salmonellosis outbreak affecting more than 16,000 persons caused by cross-contamination of pasteurized with unpasteurized milk demonstrates the potential for large-scale illness caused by the salmonellae in the food distribution system.[75]

Tick-borne Diseases

Borreliosis

Lyme arthritis, as a distinct clinical entity, was recognized as early as 1972 in residents of three communities in eastern Connecticut.[76] Lyme disease or Lyme borreliosis is now the most commonly reported arthropod-borne illness in North America and Europe. In 1981 Dr Willy Burgdorfer and colleagues first observed spirochetes in adult *Ixodes scapularis* (then called *I dammini*) ticks collected from vegetation on Shelter Island, New York, a known endemic focus of Lyme disease.[77] The bacteria were shown to react specifically with antibodies from Lyme disease patients,[77-79] and later, spirochetes were isolated from the blood of two patients with Lyme disease,[80] proving the spirochetal etiology of the infection.[78] The spirochetes were later named *Borrelia burgdorferi* (Figure 25-3), after the discoverer. The deer tick, *I scapularis*, is now known to be the primary vector of Lyme disease in the northeastern and north central United States (Figure 25-4). Other vectors are closely related ixodid ticks; including *I pacificus* in the western United States, *I ricinus* in Europe, and *I persulcatus* in Asia. Based on genotyping of bacterial isolates, *B burgdorferi* has now been subdivided into multiple *Borrelia* species or genospecies.[81] In North America, all strains belong to the first group, *B burgdorferi* sensu stricto. This species, along with two others, *B afzelii* and *B garinii*, are found in Europe, although most European disease results from the latter two. In Asia, only *B afzelii* and *B garinii* seem to be associated with the illness.[81,82] *B japonica*, which was isolated in Japan, is not known to cause human disease.[83]

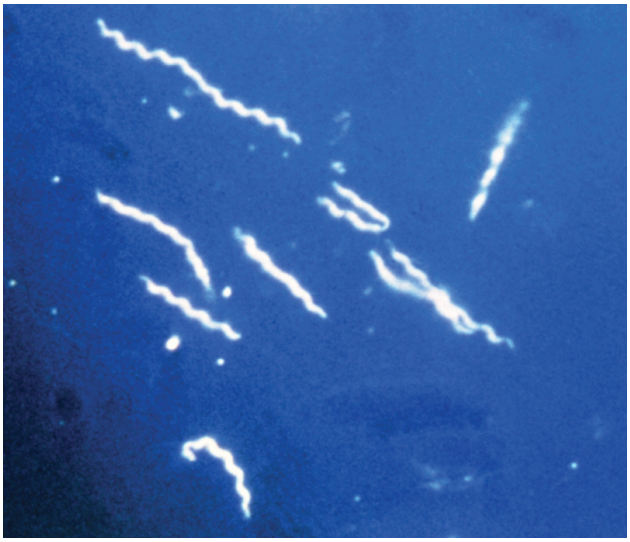

Fig. 25-3. Darkfield photomicrograph of the Lyme disease spirochete, *Borrelia burgdorferi*, magnified 400x.
Photograph: Courtesy of Centers for Disease Control and Prevention Public Health Image Library.

Fig. 25-4. *Ixodes scapularis* tick, also called the black-legged tick, is found on a wide range of hosts and is considered the main vector of the Lyme disease spirochete, *Borrelia burgdorferi*. *I scapularis* is also a vector of *Anaplasma phagocytophilum* and *Babesia microtii*, the causative agents of human granulocytic ehrlichiosis and babesiosis, respectively. Image 1669. Reproduced from: Centers for Disease Control and Prevention Public Health Image Library Web site. Photograph by Jim Gathany and provided by Michael L Levin, PhD. Available at: http://phil.CDC.gov. Accessed April 6, 2007.

Lyme disease evolves from a red macule or papule that expands annularly like a bulls-eye rash, defined as erythema migrans, which may exhibit as a single lesion or as multiple lesions. Early systemic manifestations can include malaise, fatigue, fever, headache, stiff neck, myalgia, migratory arthralgias, and lymphadenopathy, which may last for several weeks if untreated. In weeks to months after onset of erythema migrans, neurological abnormalities may develop, including facial palsy, chorea, cerebellar ataxia, motor or sensory radiculoneuritis, myelitis, and encephalitis; these symptoms fluctuate and may become chronic. Cardiac abnormalities and chronic arthritis may result.[72]

Surveillance for Lyme disease in the United States began in 1982, and it was designated a nationally notifiable disease in 1991. Since then, the number of reported cases has increased steadily, with 17,029 cases reported in 2001.[84] In 2002, 23,763 cases were reported, an increase of 40% from the previous year.[84] As with other tick-borne diseases, this continuing emergence of Lyme disease underscores the need for persons living in endemic areas to reduce their risk for infection through proper pest management, landscaping practices, repellent use, and prompt removal of ticks.

A newly recognized tick-transmitted disease that produces a rash (erythema migrans) very similar to, and often indistinguishable from, that seen in Lyme disease has been identified in the southeastern and south central United States.[85-87] Unlike Lyme disease, however, symptoms develop following the bite of the lone star tick, *Amblyomma americanum* (Figure 25-5). The disease is named southern tick-associated rash illness (STARI), but has also been referred to as Master's disease, or southern Lyme disease. *Amblyomma americanum* ticks are not known to be competent vectors of *B burgdorferi*, and serologic testing for Lyme disease in STARI patients is typically negative, despite microscopic evidence of spirochetes in biopsy samples. This finding led to speculation among physicians and researchers that a new tick-associated spirochete may be responsible. Subsequently, molecular evidence of a novel *Borrelia* species was reported from *A americanum* ticks, from white-tailed deer, and from the skin of a patient with STARI.[88-91] The organism, named *Borrelia lonestari*, was initially described only by polymerase chain reaction amplification of the flagellin B gene and 16S ribosomal DNA,[92] but has now been isolated in culture and more extensively studied.[93]

Fig. 25-5. A female lone star tick, *Amblyomma americanum*, found throughout the southeastern United States. These ticks are considered the main vectors of *Ehrlichia chaffeensis* and *Borrelia lonestari*, the agents of human monocytotropic ehrlichiosis and southern tick-associated rash illness, respectively. 2003. Image 4407.
Reproduced from: Centers for Disease Control and Prevention Public Health Image Library Web site. Photograph by James Gathany and provided by Michael L Levin, PhD. Available at: http://phil.CDC.gov. Accessed April 6, 2007.

Still other species of *Borrelia* known to cause relapsing fever are transmitted by ticks or lice. Relapsing fever caused by the spirochete *B recurrentis* can be transmitted by the body louse *Pediculus humanus*. *B hermsii,* the causative agent of tick-borne relapsing fever, is transmitted by the soft tick *Ornithodoros hermsii*.[94] The disease results in fever lasting 2 to 9 days with 1 to 10 relapses. Although the total duration of louse-borne disease usually averages 13 to 16 days, the tick-borne disease often lasts longer. Gastrointestinal and respiratory involvement is common. Neuropsychiatric symptoms also have been known to occur.[72] Relapsing fever, first reported in the United States in 1915,[95] normally occurs in the higher elevations of the western United States and southern British Columbia, Canada. After a relapsing fever outbreak among five persons visiting a cabin in western Montana,[94] spirochetes isolated from two of the patients were identified as *B hermsii,* and *O hermsi* ticks were collected from the cabin in which the patients had slept. This was the first report of both *B hermsii* and *O hermsi* in Montana, suggesting the risk of infection may be expanding beyond the previously recognized geographic range.

Ehrlichiosis

Human granulocytic ehrlichiosis is caused by infection with *Anaplasma phagocytophilum*, whereas the agent of human monocytotropic ehrlichiosis is *Ehrlichia chaffeensis*. Monocytotropic ehrlichiosis occurs in rural and suburban areas south of New Jersey to Kansas and in California, and granulocytic ehrlichiosis occurs in areas where Lyme disease is endemic.[72] The *Amblyomma americanum* tick (see Figure 25-5) transmits *E chafeensis*, and *I scapularis* (see Figure 25-4), the Lyme disease vector, also transmits *A phagocytophilum*. A spectrum of mild to severe, life-threatening, or fatal disease (< 1% mortality) occurs with ehrlichiosis. About 20% of patients have meningoencephalitis. Infection with *A phagocytophilum*

is characterized by acute and often self-limited fever, malaise, myalgia, thrombocytopenia, leucopenia, and increased hepatic transaminases.[72]

Because the *I scapularis* tick is the vector for transmission of *B burgdorferi*, *A phagocytophilum*, and *B microti*, coinfections of Lyme disease, ehrlichiosis, and babesiosis (caused by the protozoan *Babesia microtii*) can be transmitted by a bite from this tick. Ticks of the *Ixodes* genus can transmit all of these diseases as well as tick-borne encephalitis.[72] Coinfections with babesiosis and Lyme disease are known to sometimes increase the severity of both diseases.[72]

Emerging Antibiotic Resistance

Antimicrobial resistance is not a new phenomenon. Sulfonamide-resistant *Streptococcus pyogenes* emerged in military hospitals in the 1930s, and penicillin-resistant *Staphylococcus aureus* appeared in London civilian hospitals soon after the introduction of penicillin in the 1940s.[96] However, the number of resistant organisms, the geographic regions affected by drug resistance, and the number of bacterial species that are multidrug resistant is increasing. Since the 1980s, a reemergence of tuberculosis has occurred that often results from drug-resistant *Mycobacterium tuberculosis*[97] and requires the use of several (sometimes six to seven) different drugs to treat.[98] Other notable examples of multidrug resistant strains worldwide include *Enterococcus faecium*, *Enterobacter cloacae*, *Klebsiella pneumoniae*, *S aureus*, *Acinetobacter baumanii,* and *P aeruginosa*.[96] In developing countries, multidrug resistant enteric bacteria such as *Salmonella enteritidis*, *Shigella flexneri,* and *V cholerae* are major threats to public health.

Salmonella antibiotic resistance has emerged as a serious concern in agriculture as well as patient management.[99-101] Antibiotic resistance in *E coli* O157:H7 has been shown to occur rapidly following exposure to various antibiotics, including triclosan, chloramphenicol, erythromycin, imipenem, tetracycline, and trimethoprim, as well as to some biocides.[102]

Few antibiotics are more potent than vancomycin. The emergence of microbial vancomycin resistance has been of increasing concern to clinicians and public health professionals during the past decade, and surveillance systems have been instituted to monitor these pathogens.[103] *Staphylococcus aureus* is an important cause of illness and death, accounting for about one fifth of bacteremia cases in the United States.[104] The discovery of vancomycin resistance in *S aureus* clinical isolates in the United States could portend the end of the antibiotic era in medicine.[105,106]

Both hospital and home healthcare patients are significantly affected by the growing emergence of antibiotic resistance.[107,108] Restrictive guidelines have therefore been developed for the use of vancomycin and other glycopeptide antimicrobials. These guidelines include a recommendation against the routine use of vancomycin as perioperative antibiotic prophylaxis for surgical site infections.[109] Vancomycin-intermediate resistance among *S aureus* has also been identified, and subsequent guidance has been developed for its identification and control of transmission.[110] Appropriate antibiotic use will continue to be an important issue for clinicians and epidemiologists for the foreseeable future.[111]

EMERGING VIRAL DISEASES

Avian Influenza and the Threat of Pandemic Influenza

Influenza is a highly contagious, acute respiratory illness caused by one of the oldest viruses known, with clear evidence of disease dating back to the Middle Ages and probably occurring as early as ancient Greece and Rome. The virus, a member of the *Orthomyxoviridae* family, contains a segmented negative-sense RNA genome, with each segment corresponding to a gene. The segmented nature of the genome allows for the reassortment or exchange of segments (and genes) between two virus strains coinfecting the same cell. Thus, by their very nature, influenza viruses are constantly reemerging through changes in their genetic make-up. Influenza virus strains that cause pandemics are classical examples of emerging viruses. There are three main types of influenza viruses, termed influenza A, B, and C; however, only influenza A has been associated with human influenza pandemics. Two genes of special importance encode for the surface proteins hemagglutinin (HA) and neuraminidase (NA). These proteins, seen as spikes in electron micrographs (Figure 25-6), are major antigens of the virus and are involved with the interactions between the virus and host cells. Because of their importance, subtypes of influenza A viruses are often designated by their particular HA and NA types (to date, distinct hemagglutinin subtypes of influenza B and C viruses have not been observed). There are 15 HA and 9 NA subtypes, with each subtype differing by 30% or more in amino acid sequence homology.[112] All of these subtypes are found in wild waterfowl, which act as the reservoir host for influenza A viruses. Thus far, only viruses carrying one of three HA subtypes (H1, H2, H3) have crossed species barriers and established themselves in humans (H7 and H9 subtype viruses

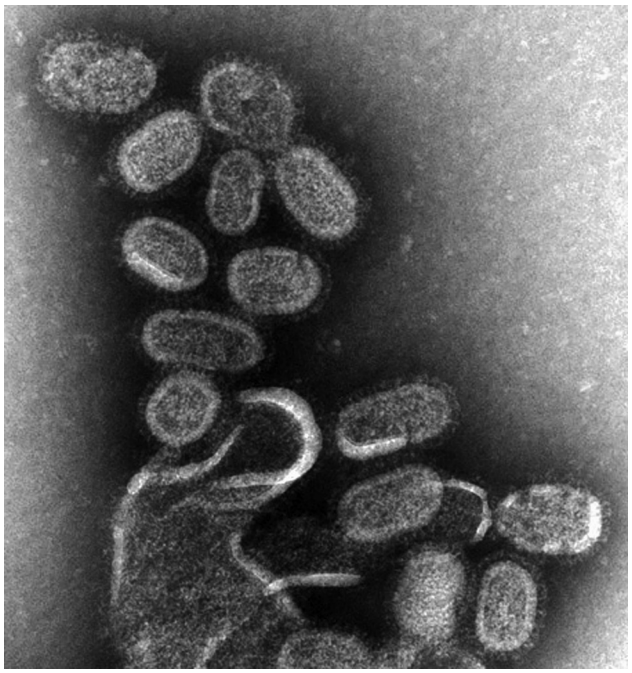

Fig. 25-6. Negative-stained transmission electron micrograph showing the reconstructed 1918 influenza virons that were collected from the supernatants of virus-infected Madin-Darby canine kidney cell culture 18-hours postinfection. Surface spikes (hemagglutinin and neuraminidase) can be clearly seen extending from the surface of the virons. 2005. Image 8160.
Reproduced from: Centers for Disease Control and Prevention Public Health Image Library Web site. Photograph by Cynthia Goldsmith and provided by Dr Terrence Tumpey. Available at: http://phil.CDC.gov. Accessed April 6, 2007.

have caused human infections, although rarely). For example, one circulating influenza strain, designated subtype H3N2, has been the most commonly isolated strain during the past 36 years.

Variants of influenza A viruses can result from mutation in the HA and NA genes. One type of variation, called antigenic drift, occurs as a result of accumulation of point mutations in the genes encoding HA and NA proteins. These point mutations, which occur randomly as the virus is copied in infected cells, are largely responsible for the annual epidemics of influenza seen during the winter months. Another type of viral change is antigenic shift, which results from the reassortment of genes that occurs when two different influenza viruses infect the same host cell. This phenomenon results in the emergence of new pandemic influenza A strains. Since 1933, when the virus was first isolated (an H1N1 subtype), major antigenic shifts (and pandemics) have occurred in 1957 ("Asian influenza," an H2N2 subtype) and in 1968 ("Hong Kong influenza," an H3N2 subtype). After a hiatus of more than 20 years, the H1N1 subtype virus reappeared in 1977. That year it did not result in severe disease, however, most likely because of the immunity of persons over 20 years of age who had been infected with the virus earlier in the century. It is highly unlikely that this virus was maintained in an animal host for over 20 years without changes; possibly, the virus was maintained in a freezer until it was somehow reintroduced into the human population. Retrospective seroepidemiological analysis can provide indications of the virus subtypes circulating during epidemics and pandemics that occurred before 1933. For instance, the 1889–1890 influenza epidemic was caused by a virus antigenically similar to the 1957 Asian strains (H2N2).[113] Likewise, the epidemic of 1900 may have been caused by a virus with an HA similar to the H3N2 pandemic virus of 1968.

Of the three influenza pandemics that occurred in the 20th century, the pandemic of 1918–1919 was the most devastating, causing an estimated 20 million to 40 million deaths worldwide. Unusually, healthy young adults between 20 and 40 years of age accounted for almost half of the influenza deaths during this pandemic. The epidemic spread rapidly, moving around the globe in less than 6 months. A reemergent 1918-like influenza virus would have even more devastating effects in today's era of rapid jet transportation and overpopulation. The pandemic killed an estimated 675,000 Americans, including 43,000 servicemen mobilized for World War I (Figures 25-7 and 25-8), and may have played a role in ending the war.[114] Its impact

Fig. 25-7. Emergency hospital during the 1918 influenza pandemic, Camp Fuston, Kansas. NCP 1603.
Photograph: Courtesy of the Otis Historical Archives, National Museum of Health and Medicine, Washington, DC.

was so profound that the average US life expectancy temporarily declined by over 10 years.[115]

Analysis of survivor antibody titers from the late 1930s suggested that the 1918 strain was an H1N1 subtype closely related to classic swine influenza virus.[116] This identification was confirmed by researchers at the Armed Forces Institute of Pathology in Washington, DC, who analyzed influenza viral RNA obtained from preserved lung tissue of US servicemen who died during the 1918 pandemic.[117] Since the original work on the HA gene, several other 1918 influenza virus genes have been sequenced and characterized.[118-121] Unfortunately, no obvious genetic changes were observed in any of these gene sequences that would account for the exceptional virulence of the pandemic virus.[122,123] However, the recent solving of the crystal structure of the HA protein derived from reassembly of extinct 1918 influenza virus may help explain the mystery.[124,125] For instance, although the 1918 virus' HA protein is distinctly avian in structure, particularly within the receptor binding site, it is able to form structural conformations that bind to human cells. This may explain how the virus could have been so virulent (because of the avian-like structure of its HA protein) and, at the same time, spread through the human population with such ease. In addition, in 2005, a team of researchers succeeded in reconstructing the 1918 pandemic virus by using gene sequences obtained from a 1918 victim (see Figure 25-6). The reconstructed virus was highly virulent, killing mice more quickly than any other human influenza virus known.[126] Such research efforts may shed more light on the highly virulent nature of the 1918 virus and help in the development of vaccines and treatments for future pandemic influenza viruses.

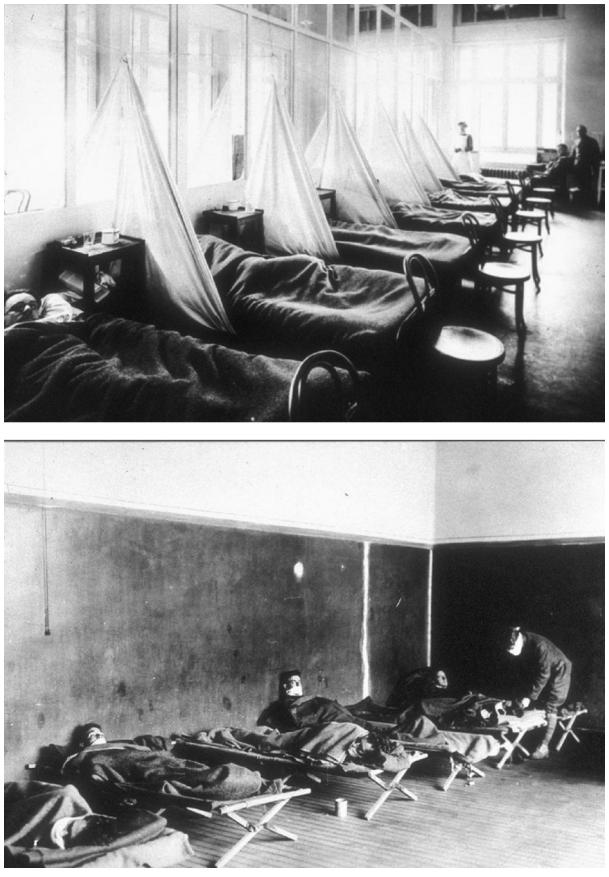

Fig. 25-8. Influenza wards, US Army camp hospitals at (**a**) Aix-Les-Bains, France (Reeve 14682), and (**b**) Hollerich, Luxembourg (Reeve 15183).
Photographs: Courtesy of the Otis Historical Archives, National Museum of Health and Medicine, Washington, DC.

Wild aquatic birds, the reservoirs of all subtypes of influenza A virus, are generally unharmed by the virus. It had been thought that these purely avian influenza viruses, although highly pathogenic for domestic poultry, did not replicate efficiently or cause disease in humans. Before the late 1990s, there were only three reported isolations of avian influenza viruses from humans. The first was from a patient with hepatitis in 1959.[127] The other two were cases of conjunctivitis, one of which was in a laboratory worker in Australia who developed infection after accidental exposure directly in the eye,[128] and the second in an animal handler who had direct contact with an infected seal.[129] All of these cases were associated with H7N7 subtype viruses. In contrast to the rarity of H7N7 avian viral isolations from humans, serosurveys of farmers in rural southern China suggest that many other subtypes of avian viruses have crossed the species barrier and infected humans.[130] Specifically, seroprevalence levels of 2% to 7% for H5 viruses alone have been reported,[130] and the seropositivity of human sera for H7, H10, and H11 viruses was estimated to be as high as 38%, 17%, and 15%, respectively.[130] It has long been believed that avian viruses could not efficiently infect humans because of receptor specificity, preventing the emergence of new pandemic strains via direct avian-to-human transmission. Transmission from aquatic birds to humans was hypothesized to require infection of an intermediate host, such as a pig, that has both human-specific and avian-specific receptors on its respiratory epithelium. Pigs were considered "mixing vessels," allowing for the reassortment between avian and human influenza viruses to occur.

However, human cases of avian influenza have recently become increasingly frequent. In 1996 an H7N7 virus was isolated from a woman who kept ducks and had conjunctivitis in her eye.[131] The source

of the virus was considered to be waterfowl because she tended a collection of 26 ducks of various breeds that mixed freely with wild waterfowl on a small lake. In the spring of 1997, an H5N1 virus was isolated from a 3-year-old boy who died in Hong Kong.[132] By the end of the same year, a total of 18 people were infected with the same H5N1 virus, and six died. Genetic analysis of these viruses showed that all of the viral genes were of avian origin (ie, they were not reassortants), and epidemiological evidence strongly suggested that direct contact with infected poultry was the route of transmission.[133,134] In addition, they appeared to be identical to viruses first isolated from an outbreak in chickens in Hong Kong earlier that same year. Because human populations lacked immunity to the H5 influenza virus subtype, there was great concern about the possibility of a major pandemic from this newly emergent virus. Fortunately, however, prompt and thorough culling of poultry on affected farms throughout Hong Kong stopped the outbreak in poultry, and enforcement of personal protection procedures for poultry handlers stopped the transmission of the novel virus to humans. In addition, the lack of evidence for human-to-human transmission in the majority of cases in Hong Kong suggested that the virus had not fully adapted to its human host.

In 2003 an H5N1 virus was isolated again in Hong Kong from a father and son who presented with respiratory illness after returning from mainland China.[135] A daughter and the mother of this family also became ill, and the daughter died while visiting mainland China. The father ultimately died of viral pneumonia, although the boy eventually recovered. Meanwhile, in Europe, outbreaks of highly pathogenic H7N7 viruses on poultry farms in the Netherlands resulted in the culling of over 30 million chickens before the virus was contained.[136] In addition, some 450 people had reported health complaints, including conjunctivitis and influenza-like illness, and a veterinarian who visited one of the farms developed high fever and severe headache, and died of respiratory distress syndrome 15 days later.[136]

Since late 2003 outbreaks of an Asian strain of highly pathogenic avian influenza (H5N1) have caused lethal illness among poultry throughout southeast and central Asia.[137] Most of these countries were experiencing highly pathogenic avian influenza for the first time. By the end of 2005, the outbreak resulted in 132 reported human cases, 68 of which were fatal.[138] In 2005 the range of the virus extended out of Asia and into Europe, with several human infections in Turkey, causing concern that a new virus subtype with pandemic potential could emerge.

Severe Acute Respiratory Syndrome

SARS is a new infectious disease that first emerged in Guangdong province of China in November 2002. Initially referred to as "infectious atypical pneumonia" by Chinese clinicians, SARS was later provided a case definition and its current name by the World Health Organization. The disease usually began with high fever and mild respiratory distress, but rapidly progressed to pneumonia within a few days. By January 2003 the disease had spread to Guangzhou, the capital of Guangdong province, and caused major outbreaks, primarily affecting healthcare workers. In February 2003, a physician from Guangdong spent a single day in a hotel in Hong Kong, where he transmitted the infection to 16 other guests. These individuals quickly spread the disease in Hong Kong, Singapore, Vietnam, and Toronto.[139] Within weeks, SARS had spread to affect thousands of people in 25 countries across five continents. By the end of the global outbreak in July 2003, there were over 8,000 recorded cases, with 744 fatalities.[140] By the end of March 2003, a novel coronavirus (SARS-CoV) was identified as the infectious agent of the syndrome.[141-143] Although researchers in China observed coronavirus-like particles in cultures grown from patient samples from Guangdong in mid-February, Chinese officials at the time reported that a *Chlamydia* bacterium caused the disease, and the coronavirus results were not reported.[144]

Where did the SARS-CoV originate and how did it become a highly lethal human pathogen? The exact origin of the SARS-CoV is still a mystery; however, the disease probably first emerged in Guangdong around November 2002.[145,146] One of the first identified SARS patients was a chef from Heyuan who worked at a restaurant in Shenzhen. As a chef, he came into regular contact with several types of live animals used as exotic game food. This prompted speculation that SARS might be a zoonotic disease. Guangdong province is famous for its "wet markets," where a wide variety of vertebrate and invertebrate animals are housed together and sold for their medicinal properties or culinary potential.[7] More than one third of the early SARS cases were among food handlers.[147] Studies with avian influenza viruses in live poultry markets have shown that such viruses amplify within the setting of a market trading in live birds.[148] Lack of serologic evidence of previous infection in healthy humans suggested that SARS-CoV had recently emerged in the human population and that animal-to-human interspecies transmission might be a reasonable explanation for its emergence. Further support for a zoonotic origin of SARS came from the initial isolation of a SARS-like coronavirus from Himalayan palm civets (Figure 25-9) found in a live animal market

Fig. 25-9. The masked palm civet was originally implicated as the possible animal source for the SARS coronavirus after SARS-like coronaviruses were isolated from animals found in a live animal market in Guangdong, China. These animals are trapped and butchered for food in southern China. This photograph was taken at a wet market in Guangzhou in May 2003.
SARS: severe acute respiratory syndrome
Photograph: Courtesy of Dr Meirion Evans, Cardiff University, United Kingdom.

in Guangdong, China.[149] However, subsequent surveys failed to find the virus in either farmed or wild civets, suggesting the civet may have served only as an amplification host for the virus. In 2005 two research teams independently identified the Chinese horseshoe bat (*Rhinolophus sinicus*) as the natural viral reservoir from which the SARS coronavirus that infected humans likely emerged.[150,151] Many people in Asia eat bats or use their feces for medicinal purposes. The researchers speculate that bats may have first passed the viruses to animals in the wild or in the live animal markets of southern China where bats are sold as food.

Emerging Paramyxoviruses

Hendra Virus

In 1994 a new member of the paramyxoviruses emerged in Brisbane, Australia, killing 14 race horses and a horse trainer.[152,153] Another worker at the stable survived with an influenza-like illness. One year later, a farmer from Mackay (800 km north of Brisbane) died as a result of encephalitis caused by this novel virus.[154] Two of his horses were subsequently shown to have died from the same virus 13 months earlier. Genetic analysis of the virus showed it was distantly related to the morbilliviruses, which contain other members such as rinderpest, measles, and canine distemper viruses. The virus was therefore initially named equine morbillivirus,[155] but was later renamed Hendra virus after the Brisbane suburb where the outbreak occurred. Serologic[156] and other evidence of infection was found in several species of Australian flying foxes (ie, fruit bats of the genus *Pteropus*) (Figure 25-10), supporting epidemiological evidence that fruit bats are the natural reservoir for Hendra virus. Field, experimental, and molecular investigations indicate that Hendra virus is an endemic fruit bat virus that has probably coevolved with its pteropid hosts.[157-159]

Additional occurrences of Hendra virus have been rare, sporadic, and limited to horses. In 1999 a horse from near Cairns in northern Queensland died from Hendra disease,[160,161] and in 2004 Hendra virus was confirmed in another dead horse from Townsville, also in northern Queensland.

Nipah Virus

Nearly 5 years after the discovery of the Hendra virus, a massive outbreak of porcine respiratory disease in Malaysia caused the deaths of 105 pig farm or abattoir workers and the eventual culling of over 1 million pigs, leading to the discovery of a new virus closely related to Hendra, called Nipah virus.[162] The predominant clinical syndrome in humans was encephalitic (unlike the respiratory syndrome seen in the infected pigs), with clinical signs including fever, headache, myalgia, drowsiness, and disorientation, sometimes leading to coma within 48 hours.[163,164] The majority of human cases included a history of direct contact with infected pigs; most were among pig farmers. Preliminary research on the new virus revealed ultrastructural, antigenic, serologic, and molecular characteristics similar to Hendra virus.[162] Follow-up molecular studies showed the genome of Nipah virus to be highly homologous to that of Hendra virus, with specific genes having nucleotide homologies between 70% and 88%, and amino acid homologies ranging from 67% to 92%.[165] Given the degree of similarity and other unique features of these viruses, both were placed in a new genus, *Henipavirus*, within the family *Paramyxoviridae*.[166] Because of the similarities between Nipah and Hendra viruses, attention focused on Malaysian bats as the source of the infection in pigs.[157] Initial surveillance efforts identified the presence of neutralizing antibodies to Nipah virus in the sera of 21 bats from five species (four species of fruit bat, including two flying fox species, and one insectivorous bat species).[167] Although no virus was isolated or viral RNA amplified from these seropositive bats, later attempts proved successful, and virus was isolated from pooled urine samples collected from a

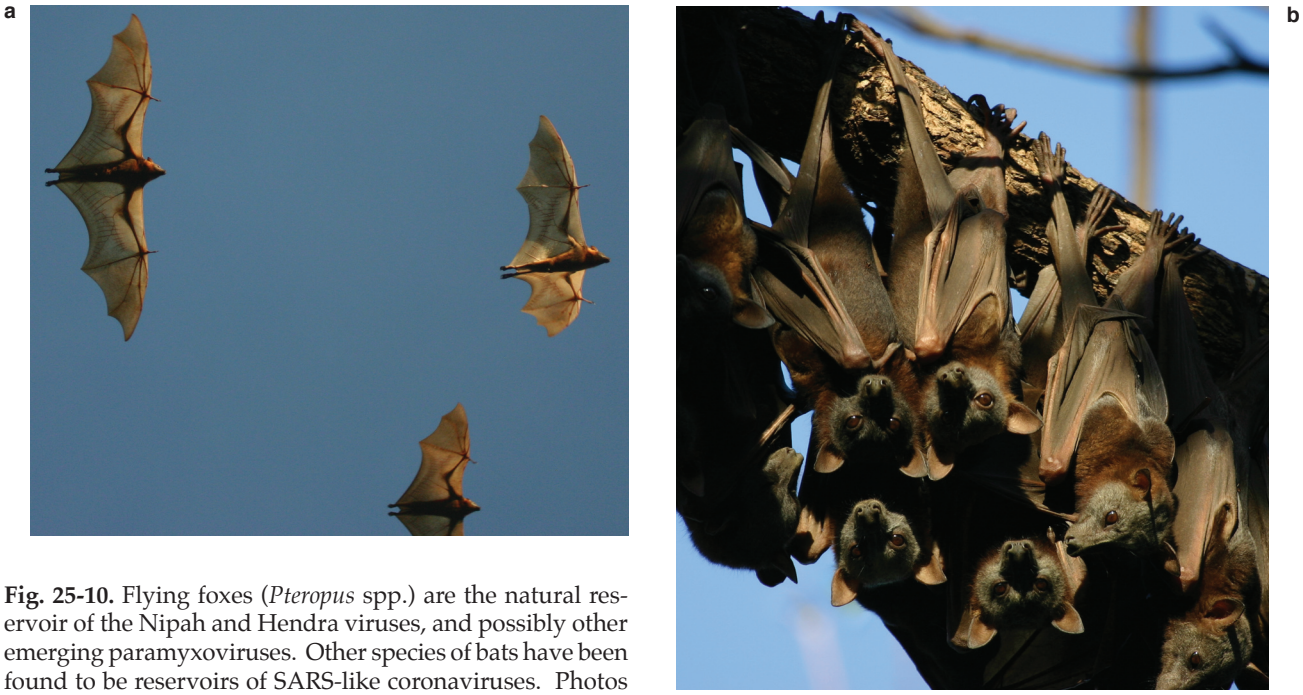

Fig. 25-10. Flying foxes (*Pteropus* spp.) are the natural reservoir of the Nipah and Hendra viruses, and possibly other emerging paramyxoviruses. Other species of bats have been found to be reservoirs of SARS-like coronaviruses. Photos show the little red flying fox (*Pteropus scapulatus*) in flight (**a**) and roosting (**b**).
Photographs: Courtesy of Raina Plowright, Department of Veterinary Medicine and Epidemiology, University of California, Davis, California.

colony of seropositive flying foxes from Tioman Island off the coast of Malaysia.[168]

The virus reemerged in Bangladesh in two separate outbreaks in 2001 and 2003, each resulting in a cluster of febrile neurological illnesses with nine and eight reported deaths, respectively.[169] In contrast to the outbreaks in Malaysia, where animal illnesses were reported and close contact with pigs was strongly associated with Nipah virus infection, no obvious zoonotic source was identified in Bangladesh. However, antibodies to Nipah virus were detected in two local *Pteropus* bats, so inadvertent direct contact with bats or bat secretions is a possible explanation for the infection (see Figure 25-10).

Menangle and Tioman Viruses

Menangle virus is a rare, previously undescribed virus that caused a single episode of reproductive disease in pigs in a large commercial piggery near Sydney, Australia, in 1997.[170] The virus caused stillbirths with deformities and occasional abortions in the affected pigs. Affected stillborn piglets frequently had severe degeneration of the brain and spinal cord. No disease was observed in postnatal animals of any age, although over 90% of them had high titers of neutralizing antibodies to the virus. Two persons who worked with the pigs developed influenza-like illness with sudden onset of malaise, chills, fever, severe headaches, and myalgia.[171] Convalescent-phase serum samples from both patients were found to have high titers of neutralizing antibodies to the virus, and extensive serologic testing showed no evidence of any alternative cause for their symptoms. Again, fruit bats were identified as the probable source of infection.[170] A large breeding colony of gray-headed and red fruit bats was found roosting within 200 meters of the affected piggery, and serum samples collected from these bats were positive for neutralizing antibodies against Menangle virus.[170]

During the search for the natural host of Nipah virus, another new member of the Paramyxoviridae family, Tioman virus, was isolated from the urine of flying foxes found on Tioman Island.[172] Nucleotide sequence and phylogenetic analysis indicate that Tioman and Menangle viruses are closely related; however, the potential of Tioman virus to cause disease in animal and humans is unknown.

Emerging Arthropod-borne Viruses: Dengue and West Nile Viruses

Mosquito-borne viruses are members of the more general category of arthropod-borne viruses or arboviruses. Human infection with arboviruses can be

asymptomatic or can cause diseases ranging from a mild febrile illness to encephalitis or even severe hemorrhagic fever in some cases. Still other infections are known to cause rash and/or epidemic arthralgia. Most arboviruses require a reservoir host such as a bird or small mammal, while using a vector, usually a mosquito or tick, for transmission to another host.[173] Because of this complex life cycle, many arboviruses are restricted to specific geographical regions. For example, Ross River and Murray Valley encephalitis viruses are restricted to Australia and surrounding islands; whereas O'nyong-nyong virus occurs only in Africa. However, because of various ecological or environmental changes (whether natural or manmade) that lead to changes in the mosquito vector distribution or genetic changes in the viruses themselves, some arboviruses may not stay within their previously known geographical regions.

Dengue Virus

Dengue is caused by one of four viral subtypes (designated DENV-1 to DENV-4) and is one of the most common mosquito-borne viral infections of humans, with up to 100 million cases reported annually and some 2.5 billion people living at risk of infection in tropical and subtropical regions of Africa, Asia, and the Americas.[174] Infection with dengue virus can present in several clinical manifestations. Classical dengue fever is an acute febrile illness that often occurs in children, characterized by fever, severe headache and muscle aches, nausea, vomiting, and rash. This acute illness usually lasts for 8 to 10 days and is rarely fatal. A more severe form of dengue infection is dengue hemorrhagic fever/dengue shock syndrome (DHF/DSS). DHF usually begins during the first week of the acute illness and can lead to hemorrhagic manifestations, including petechiae, ecchymoses, epistaxis, bleeding gums, and gastrointestinal tract bleeding.[175] DSS occurs if the patient goes on to develop hypotension and shock from plasma leakage and circulatory failure. This happens in about one third of severe dengue cases (especially in children) and is often associated with higher mortality. Convalescence for patients with DHF is usually short and uneventful, and if shock is overcome, patients usually recover within 2 to 3 days.[175] The pathogenesis of DHF/DSS is complicated and not well understood. Two theories are frequently cited to explain the pathogenetic changes that occur in DHF/DSS. The most commonly accepted theory, known as immune enhancement,[176,177] suggests that patients experiencing a second infection with a heterologous DENV serotype have a significantly higher risk of developing DHF/DSS. Preexisting heterologous dengue antibody recognizes the infecting virus and forms an antigen-antibody complex, which is then bound to and internalized by immunoglobulin Fc receptors on macrophages. Thus, it is hypothesized that prior infection, through a process known as antibody-dependent enhancement, enhances the infection and replication of DENV in mononuclear cells.[175] The other theory assumes that dengue viruses change genetically as a result of selective pressures as they replicate in humans and/or mosquitoes and that the phenotypic expression of these genetic changes may include increased virus replication and virulence. All the data taken together suggest that a combination of age and the viral, immunopathogenic, and genetic background of the person play a role in disease severity.[174]

Although dengue viruses were first identified in southeast Asia in the 1940s and 1950s, evidence suggests that they derived from a primitive progenitor introduced to Asia from Africa about 1,000 years ago.[178] Studies of dengue virus ecology in sylvatic habitats of western Africa and Malaysia have identified transmission cycles involving nonhuman primates as reservoir hosts and arboreal, tree-hole dwelling *Aedes* species mosquitoes as vectors.[179,180] Efficient interhuman dengue virus transmission probably requires a human population of 10,000 to 1 million people, a feature of urban civilization that did not exist until about 4,000 years ago, suggesting that the sylvatic cycle is probably ancestral.[181] Further support for this idea comes from studies suggesting that a zoonotic transfer of dengue virus from sylvatic to sustained human transmission occurred between 125 and 320 years ago.[178] In the past 300 years, these viruses have become established in the urban centers of the tropics. The principal urban vector, *A aegypti*, is highly domesticated and adapted to humans, preferring to feed on people and lay their eggs in artificial containers in and around houses. *A albopictus* (the Asian tiger mosquito [Figure 28-11]) is a secondary vector of dengue viruses. Dengue occurs rarely in the United States, primarily in southern Texas. However, because the vectors are distributed throughout much of the southeastern United States, a greater potential for future emergence of dengue in the United States exists.

In the past 25 years a marked global emergence of epidemic dengue has occurred, with more frequent and larger epidemics associated with more severe disease.[175,182,183] The reasons for this are not fully understood, but are thought to stem from major demographic and societal changes over the past 50 years, particularly the unprecedented global population growth and associated unplanned and uncontrolled urbanization, especially in the tropical developing countries.[175] Other potential factors associated with the global emergence of dengue include the lack of

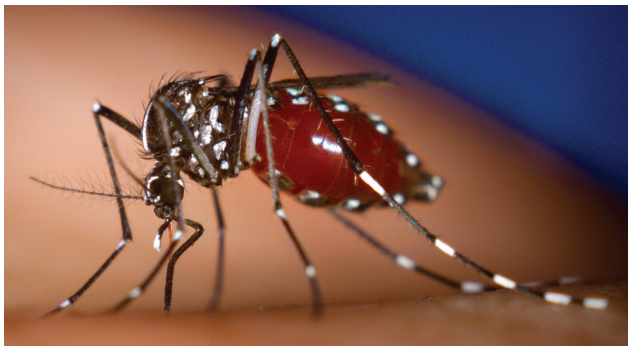

Fig. 25-11. A female *Aedes albopictus* mosquito feeding on a human host. These mosquitos, along with *A aegypti*, are competent vectors of dengue virus. 2003. Image 4490. Reproduced from: Centers for Disease Control and Prevention Public Health Image Library Web site. Photograph by James Gathany. Available at: http://phil.CDC.gov. Accessed April 6, 2007.

effective mosquito control in many tropical areas where dengue is endemic, increased international air travel, and a general decay in public health infrastructure in most countries over the past 30 years.[175]

West Nile Virus

West Nile virus (WNV) was first isolated in 1937 from the blood of a febrile patient in the West Nile district of northern Uganda. It is now one of the most widely distributed of all mosquito-borne arboviruses, found in areas throughout Africa, Europe, Asia, and North America (Figure 25-12). Yet until recently, it was completely exotic to the western hemisphere. In 1999 WNV emerged in the New York, New York, area as the cause of an outbreak of meningoencephalitis resulting in 7 deaths among 62 confirmed cases.[184] There was a concurrent outbreak among the horse population on Long Island, New York, resulting in 25 equine cases including 9 fatalities.[185] The principal mosquito vectors were likely *Culex pipiens* or other related *Culex* species; however, the virus has been isolated from a number of other mosquito species and even, in some cases, from ticks.[186,187] The virus has been shown to be capable of infecting over 50 species of mosquitoes and ticks.[187,188] Since the introduction of WNV into New York in 1999, the virus has spread across the United States (Figure 25-13). In addition, since 2000, WNV has spread into Central America, with virus being isolated in Mexico, El Salvador, and

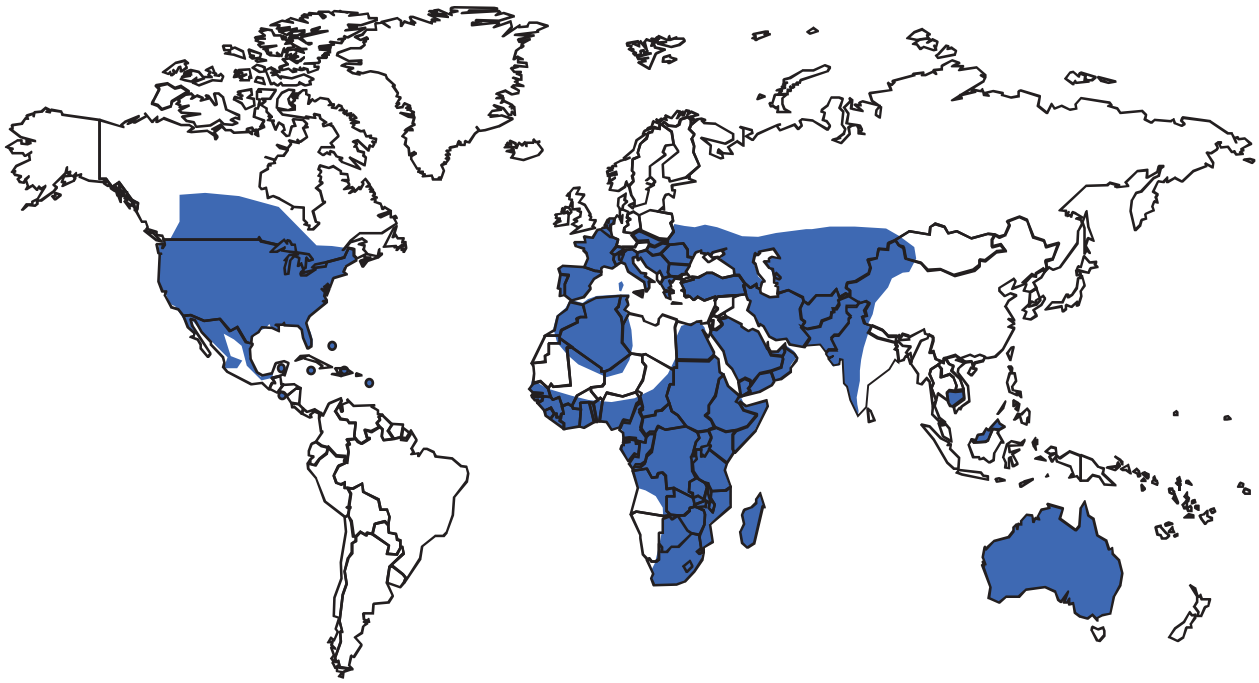

Fig. 25-12. Approximate geographic range of West Nile virus, 2004.
Map: Courtesy of Dr Robert Lanciotti, Arbovirus Diseases Branch, Centers for Disease Control and Prevention, Fort Collins, Colorado.

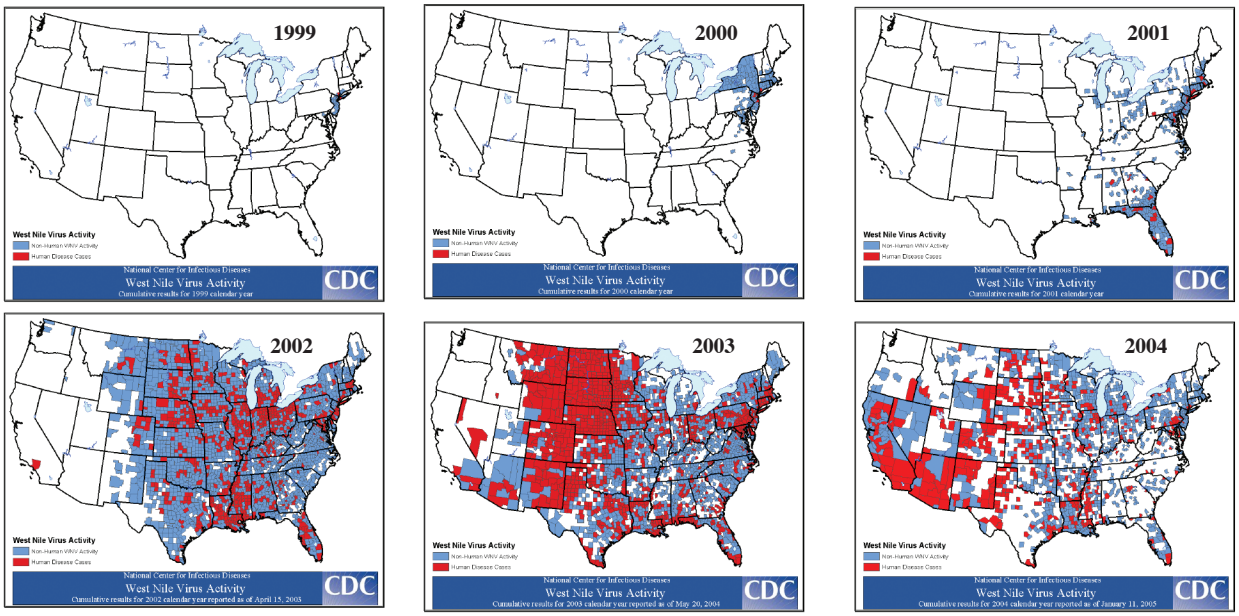

Fig. 25-13. Spread of West Nile virus activity across the United States, 1999 to 2004. Data represent nonhuman West Nile virus activity (in blue) and human disease cases (in red) in the United States by county.
Reproduced from: National Center for Infectious Diseases, Centers for Disease Control and Prevention.

the Caribbean Islands.

Recent years have seen a high incidence of human infection with WNV through blood transfusion, mother-to-fetus transmission, and transmission in breast milk and by organ transplantation, causing even greater public health concerns.[189-194]

GENETICALLY ENGINEERED THREATS

Without human intervention, the natural world has produced innumerable microbial agents that continue to emerge as new or newly observed causes of disease. Human activity has also played a huge role in the emergence of many diseases, but this role has been inadvertent, rather than deliberate. The spread of HIV, for example, can be attributed almost entirely to human behavior, and the same was true of the spread of smallpox. Historically, both microbial agents and the affected populations have tended to change during the course of disease outbreaks. In Europe, several generations of exposure to smallpox and measles ensured the survival of those most resistant to these diseases; when the diseases were introduced in the New World, unchecked contagion and decimation of the unexposed populations occurred.[195,196] A classical example of agent-host adaptation in animals was the intentional introduction of myxomatosis (an orthopoxvirus similar to smallpox that infects rabbits) into Australia in an attempt to control or eliminate a scourge of rabbits. At first, rabbit mortality was very high, but in time the rabbits acquired a degree of genetic resistance. In parallel, virulence diminished in the circulating virus, which persisted and was shed over a longer period of time in infected rabbits.[197] For both rabbit and virus, natural selection favored survival of the species. Humans have intentionally disturbed this "natural order," from using relatively benign forms of disease as vaccines against the most virulent forms (eg, variolation, or the classical adaptation of measles, mumps, and rubella vaccines) to selecting the most virulent disease agents for biological weapons programs (the latter was finally stigmatized and outlawed in the Biological Weapons Convention Treaty). Other microbial perturbations have been unintended, such as the treatment-based selection of antibiotic-resistant bacteria now widespread in hospitals.[198]

More recently, humankind has acquired the technical capacity to create microbial threats far more deadly than natural evolution could create or sustain. Genetic engineering, the intentional molecular reshuffling of genes between and among microbial agents and higher

organisms, has proven like so many technologies to have capacity for both good and ill. A few examples from the scientific literature illustrate the seriousness of the threat of genetically engineered microorganisms.

For anyone moderately skilled in microbiology, it is obvious that otherwise harmless bacteria may be engineered to synthesize toxins made by unrelated lethal strains of bacteria. Antibiotic resistant strains of *B anthracis*, the causative agent of anthrax, have been derived not only by biological selection, but also more directly by genetic engineering.[199-201] Unauthorized conduct of most such experimentation has become not only difficult but illegal, subject to fines and incarceration, in many countries including the United States.

However, skilled laboratory researchers can now easily manipulate viral genomes by recovering infectious viruses from DNA clones. The progression of this technology with human pathogens began about 20 years ago with the simpler viruses (positive-sense, single-strand viruses with small genomes), such as poliovirus,[202] alphaviruses,[203] and flaviviruses.[204] The technology has grown to include negative-strand viruses (eg, vesicular stomatitis virus, respiratory syncytial virus, Ebola virus, and Crimean-Congo hemorrhagic fever virus) and segmented viruses (eg, influenza virus). Even the relatively huge genome of vaccinia virus has yielded to artificial resuscitation from DNA cloned into bacteria.[205] In an experiment that was alarming to some observers in its simplicity, the capacity to derive a human pathogenic virus (poliovirus) by chemical synthesis was demonstrated.[206] Even more controversial are the efforts to genetically resurrect the deadly 1918 influenza virus[207-210] and the proposals to genetically manipulate smallpox virus.[211]

In addition to the potential for recovering hazardous viruses from DNA clones, risks of accidental or malevolent outcomes are further elevated with engineered recombinant viruses. Experiments designed to create or improve vaccines, to understand interactions between virus and host, or to unveil some mysteries of the viruses themselves have simultaneously proven the ease with which bioactive and sometimes harmful molecules may be inserted into viruses. A large body of work with recombinant poxviruses was considered benign until a mouse poxvirus (ectromelia virus) rendered more virulent by its modification to coexpress a molecule of the immune system (interleukin-4) was reported.[212] This result was merely part of a progression of studies of similar design and outcome,[213] but its timing (2001) crystallized the potential problem. This technology, applied to a wide array of human pathogens, remained underappreciated until federal regulators began defining and implementing safety and biosurety rules for select agents.

Ultimately, the capacity to create deadly and possibly even apocalyptic new organisms through genetic engineering is restrained largely by technical knowledge and opportunity, and also by awareness and intent. That is, techniques easily accomplished by skilled scientists are extremely difficult for the untrained and unequipped. However, a determined person with the appropriate knowledge and skills may succeed in malevolent creation of genetically engineered microorganisms. Unfortunately, such organisms could also be created by well-intentioned scientists who underestimate the unexpected consequences of their work.

What countermeasures and solutions exist? New laws and regulations to emphatically restrict accidental or intentional creation of new deadly organisms, or possession of the deadly agents existing in nature, have already been imposed in the United States (eg, Public Law 107-188[214]), but these bounds are difficult if not impossible to enforce internationally. Also helpful are the myriad coordination meetings and rehearsals for public health responses to pandemic natural threats such as smallpox or a deadly pandemic influenza virus; in the case of the outbreak of a contagious genetically engineered microorganism, classical methods of epidemiology and quarantine would likely be helpful. Also encouraging is the application of the newest technologies to both diagnostics and bioforensics, likely shortening the time in which the nature and design of a newly emerged causative agent would remain unknown. Unfortunately, development of specific medical countermeasures (vaccines, therapeutic drugs) for a previously unknown organism can take months and usually years. One response to this problem is to fund the search for generic methods of boosting innate immunity to provide increased resistance to most or all infectious agents. A related approach is to target common cellular pathways used and shared by many unrelated agents, especially viruses. Even if medical countermeasures were nominally available, however, both genetically engineered and conventional agents could cause great localized harm and widespread panic.

SUMMARY

Emerging infectious diseases are among the most important future threats facing both military and civilian populations. These are diseases caused by a variety of infectious agents (ie, bacteria, viruses, fungi, and parasites), some completely new to mankind, and others only newly recognized. Still others

may be common commensals that have acquired virulence factors (eg, toxins) or antimicrobial resistance genes though natural or unnatural (ie, genetic engineering) means.

Despite many successes in disease control and prevention in the 20th century, infectious diseases remain the leading cause of death worldwide and the third leading cause of death in the United States. AIDS, which was first recognized in 1981, is the most dramatic example of a new infectious disease that has emerged rapidly in the past 25 years. The AIDS pandemic will continue to put large numbers of people at risk for new and reemerging opportunistic infections. The rapid spread of the WNV across the United States after its introduction in 1999 and the increasing problem of antimicrobial resistance are other examples of microbes' ability to emerge, adapt, and spread.

Future threats are difficult to predict but will certainly include the increasingly complex challenges of foodborne and waterborne diseases, the threat of another influenza pandemic, emerging antibacterial and antiviral resistance, and the likelihood of increasing problems with zoonotic diseases. What new diseases will be encountered in the next 20 years? What role will the increasingly advanced field of molecular biology play? Will other infectious agents from the past, in addition to the 1918 influenza virus, be resurrected? Or will increasingly advanced bioterrorists or rogue nations be able to create the ultimate weapons though genetic engineering? Meeting these challenges will require continued research with a multidisciplinary approach, using the expertise of physicians and veterinarians trained in public health, microbiologists, pathologists, ecologists, vector biologists, and public health officials, both military and civilian.

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