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The ABCs of bioterrorism for veterinarians, focusing on Category A agents

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At present, bioterrorism is a household word. During the past 2 years, our society has changed dramatically, becoming shaped and in some measure confined by terrorism. The United States presently faces an ongoing barrage of terrorist threats, perceived terrorist threats, and security risks unmatched in history. Among the threats for which we are most concerned, the most notable is bioterrorism. In the United States, bioterrorism has become a focal point, a polestar, for intelligence gathering, threat assessment, preparedness efforts, policy making, education, and public information campaigns. The threat of biological attack is so great of a concern that it was a leading factor given by President George W. Bush for the United States to enter into war against Iraq, a country thought to be hiding biological and chemical weapons and known to have produced them in the past.[1-4]

In today's atmosphere of globalization, emerging zoonoses, and risks of bioterrorism, the importance of veterinarians and their skills in addressing these new challenges cannot be overstated. As guardians of animal and public health, veterinarians must be educated on specific details of bioterrorism and zoonotic disease agents and stand ready to address these issues for the betterment of animal and human health alike. Two recent events regarding emerging diseases have emphasized the interconnectedness, or union, that exists between animals, veterinarians, and public health. **Severe acute respiratory syndrome (SARS)** first erupted in November 2002 in China's Guangdong province[5] and quickly spread across the globe. To date, the disease has resulted in 8,437 probable human cases with 813 deaths.[6] As of July 15, 2003, there have been 344 suspected and 74 probable cases of SARS but no deaths within the United States.[7] The novel SARS-associated coronavirus identified[8,9] from these patients appears to be distantly related to known coronaviruses[10] and is speculated to have its origins in animals.[8-12] In May 2003, Chinese researchers detected the SARS coronavirus in 6 masked palm civets and 1 raccoon-dog by **polymerase chain reaction (PCR)** methods and virus isolation[12]; genome sequencing revealed 99.8% homology to the human SARS virus. One Chinese ferret badger in that investigation had neutralizing antibodies to the virus. It is noteworthy that > 33% of the patients in Guangdong Province who had onset of the disease before February 1, 2003, were food handlers (people who prepared or served food or who slaughtered, handled, or sold food animals)[11]; in that region of China, there is much opportunity for human contact with many different and exotic animal species in the markets. It is not clear whether animals such as the palm civet, raccoon-dog, or the Chinese ferret badger served as reservoirs and transmitters of this virus or if they acquired it from another source.[11] There is also evidence (from serologic testing, virus isolation, or PCR testing) that the virus infects many other animal species, such as domestic cats and dogs, bats, snakes, monkeys, and wild pigs.[12-14] To date, genetic analysis of the SARS coronavirus isolates from animals has revealed that they are identical or similar to human isolates.[12-14] Researchers have isolated the virus from pharyngeal and nasal swabs of cats and pharyngeal swabs of ferrets experimentally infected with the SARS virus.[15] Only ferrets developed clinical signs of SARS in that study. Whether SARS is zoonotic is not known, but it seems highly likely; although SARS appears contained at this time, reemergence of the disease would not be unexpected in the near future. Also, disease caused by the monkeypox virus emerged for the first time in the western hemisphere in a multistate outbreak detected in June of this year[16]; the outbreak was associated with the importation into Texas of 800 small mammals (including 762 wild rodents) from Africa. Polymerase chain reaction assays and virus isolation performed on specimens from some animals in this shipment yielded positive results for monkeypox in 6 rodents (1 Gambian rat, 3 dormice, and 2 rope squirrels).[17] To date, 71 human cases (of which 35 were confirmed via laboratory evaluations and 36 were considered suspect or probable) of monkeypox have been reported from 6 states,[17] including cases among veterinarians and veterinary staff.[18] There have been no monkeypox-associated deaths. In the 35 confirmed cases, the patients had had contact with prairie dogs that were obtained as pets and that had been in contact with African ani-

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mals.[17] Four prairie dogs tested positive for monkeypox virus by PCR and immunohistochemical assays. Of the 762 African rodents, 178 were untraceable beyond the point of entry into Texas because of a lack of records.[17] In an effort to control this outbreak, the **Centers for Disease Control and Prevention (CDC)** quickly alerted animal owners, health care providers, and veterinarians about this disease and released multiple fact sheets and guidance reports. At this time, the outbreak has been contained and there have been no human deaths.

Animals serve as sentinels for human disease[19-23]; after the intentional release of a biological agent, they may show clinical signs of disease long before signs are evident in humans. As Huxsoll et al[24] comment, there are more animals than people in the United States, and thus they should not be overlooked as disease sentinels. Therefore, not surprisingly, veterinarians may be the health care professionals to first detect a release of a biological agent and call attention to an impending outbreak; this action may, in turn, prevent catastrophic human suffering and loss of life. The monkeypox outbreak clearly illustrates how veterinary practitioners may encounter a zoonotic disease (one that might also be used for bioterrorism) that affects animals and owners and highlights the importance of early reporting of unusual and suspicious clinical signs of disease to veterinary and public health officials. Domestic animals may also act as disseminators, secondary spreaders, or shedders of disease agents, which perpetuate an outbreak and complicate control measures with the potential for spillover of the agent into wild populations (as might occur with plague or monkeypox). Veterinarians must be aware of the biological agents that are most likely to be used in a terrorist attack and have knowledge of how they might be used, their characteristics, and their zoonotic properties. More than 50 years ago, Todd[25] hinted in his publication on agroterrorism that veterinarians could be involved in issues of bioterrorism. Without doubt, veterinarians have become deeply involved. The purpose of this article is to familiarize veterinarians with the clinical signs of infection in animals and humans and the zoonotic potential of several of the microbial and viral agents most likely to be used by bioterrorists targeting humans; furthermore, an update on efforts to promote preparedness is provided. The emphasis of this article is on those agents deemed most critical to public health, that is, those designated as Category A agents by the CDC.

Terrorism

Despite current perception, terrorism is not new to the United States. Indeed, between 1980 and 1999, the Federal Bureau of Investigation recorded 327 actual or suspected incidents of terrorism in the United States and another 130 incidents that were prevented.[26] Historically, terrorism perpetrated by domestic terrorists or terrorist groups has been this nation's greatest concern. This has changed to some degree. The first experience with international terrorism on US soil began with the bombing of the World Trade Center in 1993.[27] In 2001, the people of the United States became more acutely aware of their vulnerability to international terrorism when airplanes were used as weapons to kill approximately 3,000 people.[28] Bioterrorism became apparent with the postal mailings of letters containing spores that resulted in 22 human cases of anthrax (of these 22 persons, 11 contracted the disease via inhalation and 11 via a cutaneous route); 5 patients died.[29] However, this was not the first experience of dealing with bioterrorism in the United States. In 1984, the population (approx 10,500 persons) of The Dalles, a small town in Oregon, became the target of a cult known as the Rajneeshees, which intentionally contaminated the salad bars at local restaurants and produce at a local grocery store with *Salmonella* serotype Typhimurium in an attempt to sway the outcome of an upcoming county election; this act eventually resulted in 751 people becoming ill.[3,30]

Although there are fewer international terrorist incidents than there were 15 to 20 years ago,[31] the incidents that occur presently often result in higher numbers of casualties and deaths.[27] Terrorists still rely almost entirely on conventional weapons such as bombs and guns to carry out their missions,[27] and bioterrorism and agroterrorism play a very small role overall in their activities. On review of data available to the public, there are only 6 confirmed instances worldwide in which terrorists or terrorist groups used or tried to use biological weapons.[32] Of those incidents, only 2 (the Rajneeshee attack in 1984 and the contaminated postal mailings in 2001) have resulted in human illness or death; however, other undocumented incidents also may have occurred. Historically, most attacks against humans that involved the use of biological agents have been carried out by individuals or groups with criminal intent, not terrorism, and are thus classified as biocrimes.[32]

Bioterrorism has been given many definitions by many different people and agencies, ranging from very simple to very complex. For the purposes of this article, the definition of bioterrorism adopted will be a refinement of that put forth by Carus,[32] which is, "The use, or threatened use, of microorganisms or their toxins against humans, animals, or plants by individuals or groups motivated by political, religious, ecological, or other ideological objectives."

Unlike other definitions, this definition does not try to rigidly delimit the motivations or goals behind the use of a biological agent to those that are politically or socially oriented, but instead allows for its use in a broader context. This definition now includes the use of biological agents by apocalyptic groups seeking to cause mass deaths (eg, the Aum Shinrikyo) and by groups who view a biological agent as a tool to be used to achieve a specific objective. Both these types of groups use biological agents not for their psychological effects (ie, to induce terror), but rather for their usefulness in completing a task.[32] Inherent to all forms of terrorism is the evocation of fear, panic, and dread and the targeting of noncombatants.[33]

It must be pointed out that there is a distinction between use of a biological agent as a weapon and a biological weapon. In the first instance, any microbe or its

toxin could be used in numerous ways to achieve a particular goal, such as waging terrorism to promote or change political or social policies, to wreak economic devastation, or to fulfill religious ideologies. For more self-serving ends, these agents might be used to commit common crimes such as extortion, assassination, murder, or revenge. A biological weapon, on the other hand, is more than the agent alone and implies a physical weapon created for involvement in a military tactic. A bioweapon consists of the biological agent (the payload), the munition (a container to keep the agent viable or intact during delivery), delivery mechanism (eg, missile or aircraft), and the dispersal mechanism (spray device or other means to disseminate the agent).[4]

There are several routes by which a biological agent can be disseminated (eg, in food and water or via insect vectors or direct contact); aerosolization is viewed by most experts as the most efficient and most dangerous means of targeting humans (as well as animals in many situations) and will therefore be the primary route addressed in this article.

The ABCs of Bioterrorism

There are many microorganisms that could be used for the purpose of bioterrorism. In reality, almost every pathogen can be used for terrorism provided that it is used in a manner that takes advantage of its inherent pathogenic characteristics and the process of dissemination does not reduce the pathogen's viability beyond its effectiveness. However, not all microbial agents are created equal when it comes to their effectiveness as bioterrorism agents, and some are better suited to that purpose than others. In 1999, in an effort to enhance our national public health response to acts of bioterrorism, Congress designated the CDC as the lead agency to address public health planning for such an event.[34] Experts from different fields were gathered to examine and categorize the threat posed to public health by various biological agents and toxins. Criteria used to categorize an agent according to its threat potential included the public health impact of the agent; the potential of the agent to be used against a large population; the potential for human-to-human spread; stability of the agent; public perception of the agent; ability of the agent to incite fear among the public; and public health preparedness measures required to combat it, such as the stockpiling of medications and vaccines as well as diagnostic and surveillance needs.[34] The outcome of the assessments was the grouping of the agents into 1 of 3 categories designated A, B, or C (**Appendix 1**). Most of these agents are zoonotic.

Agents in Category A

The Category A agents are those agents that have the greatest potential for inflicting large numbers of human casualties, can be manufactured and disseminated on a large scale, require significant efforts in public health preparedness, and are most familiar to the public.[34] Of the 6 agents included in this category, all have the potential to be dispersed via aerosolization, although some are more stable in the environment or require a smaller infectious dose than others.

Anthrax—Anthrax, caused by *Bacillus anthracis*, has historically been associated with workers of the wool industry who, during the 19th century, developed inhalational anthrax when working with imported contaminated wool and alpaca and goat hair; thus, the disease became known as woolsorter's disease in England,[35,36] or ragpicker's disease in Germany and Austria.[37] *Bacillus anthracis* is an organism that has the form of a bacillus in a nutrient-rich environment, such as a living body, but transforms into a hardy spore (that is capable of surviving decades) when exposed to air and when nutrients are depleted.[38] Anthrax is by far the most recognizable of the diseases associated with bioterrorism agents and is perhaps the most feared. In 2001, the detection of postal mailings that contained spores of *B anthracis* forced many people to realize that bioterrorism is a real threat and that an effective attack can be carried out via simple means. The incident also demonstrated the effectiveness of aerosolization in dispersing this agent.

Humans typically develop anthrax from contact with infected animals or contaminated animal products.[38,39] There are 3 clinical forms of anthrax in humans: cutaneous, gastrointestinal, and inhalational. The cutaneous form accounts for 95% of all human cases worldwide.[40] From 1944 to 1994, there were 224 cases of cutaneous anthrax in the United States[38]; naturally occurring cutaneous anthrax was last reported in August 2000 in an inhabitant of North Dakota in association with a large anthrax outbreak in livestock.[41] Following skin inoculation with the organism, the spores germinate, producing toxin and resulting in localized edema. A pruritic macule or papule will ulcerate, leading to development of a black eschar that is usually associated with considerable edema. The eschar typically falls off 1 to 2 weeks later.[38] The gastrointestinal form of anthrax is primarily associated with the consumption of contaminated, poorly cooked meat. The upper or lower portions of the gastrointestinal tract can be affected. With the oropharyngeal form, signs include oral or esophageal ulcers, lymphadenopathy, sore throat, edema, possible respiratory distress due to massive edema, and potentially sepsis.[38,42] Signs associated with the lower portion of the gastrointestinal tract include intestinal ulcerations, fever, nausea, vomiting, severe bloody diarrhea, and acute abdominal pain.[38,42] Inhalational anthrax has 2 stages. The initial phase is characterized by nonspecific signs such as fever, malaise, myalgia, cough, some thoracic or abdominal pain, chills, headache, vomiting, and dyspnea.[38,42] The second phase begins abruptly a few days later with continued fever, acute dyspnea, diaphoresis, cyanosis, and shock. Stridor may be present as well. Mediastinal widening may be detected via thoracic radiography.[38,42] Case-fatality rates can be as high as 20% for cutaneous anthrax,[38,41,42] 50% for gastrointestinal anthrax,[42] and 95% for inhalational anthrax.[43] In the United Sates, naturally occurring inhalational anthrax is rare[39]; only 18 cases were reported from 1900 to 1976,[39,41] the last of which involved a home craftsman from California who worked with contaminated yarn.[37,44]

Experimental trials in monkeys involving the aerosolization of *B anthracis* spores demonstrated that fatal disease can develop as late as 58[45] to 98 days[46] after exposure.[38,46] For this reason, administration of ciprofloxacin for 60 days was recommended as treatment for those persons exposed to *B anthracis* spores via postal mailings in 2001,[47] although the US Department of Health and Human Services later offered prophylactic antimicrobial treatment for an additional 40 days to those individuals.[48] Previously, the number of *B anthracis* spores required to kill 50% of people exposed to them (LD_{50}) in humans was estimated to be 2,500 to 55,000[38]; however, recent extrapolation by Peters et al[49] suggests as few as 1 to 3 spores may cause infection.

Veterinarians assumed a wide range of roles and responsibilities in the anthrax investigation of 2001. Of the 22 cases of anthrax that were detected,[29] the 11th person to develop inhalational anthrax was a 94-year-old woman from Connecticut. In an effort to better identify previously undetected cases (human or animal) in the Connecticut area, surveillance was expanded to include veterinary clinics.[50] Surveillance included 140 veterinary practices (with 365 veterinarians), representing 48% of all licensed practitioners in Connecticut. Animal deaths for which the cause was not diagnosed were reported at 18 of the 140 practices, but anthrax-related deaths were not reported at any of the practices.[50]

Within the United States, hyperendemic areas for anthrax include South Dakota, Oklahoma, Nebraska, and parts of southwestern Texas,[40] although the disease in animals had been reported in almost all states by the end of the 20th century.[51] Anthrax affects livestock primarily, of which the most susceptible are cattle, followed by sheep, horses, and goats (in decreasing order of susceptibility).[51] Of the 3 forms of anthrax in animals (ie, peracute, acute, and subacute or chronic), the peracute form of the infection is evident in most affected ruminants and often results in sudden death.[51] In animals that die suddenly, hemorrhage from the anus, nostrils, vulva, mouth, or eyes may be observed, in addition to incomplete rigor mortis and bloating. Clinical signs in cattle and sheep with acute disease include signs of depression, listlessness, fever, labored breathing, and abortion; also, swelling of the tongue and in regions of the throat, sternum, perineum, and flanks may develop.[51,52] Horses with acute disease may develop colic and die within 48 to 96 hours.[52] Pigs may develop any of the 3 clinical forms of anthrax, including pulmonary disease, which occasionally develops in neonatal pigs after inhalation of contaminated dust.[52]

Overall, pigs, dogs, and cats are more resistant to *B anthracis* infection than livestock,[51] and reports of anthrax in these species in the United States are rare. Dogs and cats are typically infected through the consumption of contaminated raw meat, resulting in clinical signs associated with the upper portions of the gastrointestinal tract including edema, local inflammation, and necrosis.[53] Swelling of the head, neck, and throat region also develops. Unusual presentations can occur, such as the reported case of anthrax in a dog that was associated with signs of hemorrhage in the distal portions of the gastrointestinal tract, splenomegaly, and suspected cutaneous lesions.[54] In an effort to better define susceptibility and clinical disease, Gleiser et al[55] experimentally exposed 14 dogs and 14 pigs to aerosolized anthrax spores. The only clinical evidence of infection was that of fever in 3 dogs and 8 pigs. Pulmonary lesions were detected post-mortem in 3 dogs and 2 pigs, but none of the animals developed systemic signs of anthrax, and none died from infection before euthanasia.

Veterinarians should not perform a necropsy on an animal suspected to have died as a result of anthrax because vegetative bacteria within the carcass will sporulate and contaminate the surrounding area.[51] It is instructive to note that from 1957 to 1971, 6 veterinarians contracted cutaneous anthrax after conducting necropsies on infected animals.[56] If the carcass remains intact, *B anthracis* organisms will be killed as a result of the putrefactive process after a few days, although some bacilli may escape in fluids draining from body openings during or after death and contaminate the local environment.[40,51] It is recommended that disposal of anthrax-affected carcasses involve either burial (with a layer of quicklime deposited over the carcass) or burning.[51]

Biological attack scenario involving *B anthracis*

A hypothetical release of *B anthracis* in a city of 100,000 inhabitants, without intervention, under optimal meteorological conditions, could result in 50,000 illnesses and 32,875 deaths with a case-fatality rate of 50% to 80% depending on the incubation period.[57] Costs associated with the aftermath of this type of bioterrorist attack could near $26.2 billion.[57] In 1970, the **World Health Organization (WHO)** estimated that under specific conditions, 50 kg of dried *B anthracis* spores released near a developed city of 5 million inhabitants would result in 250,000 illness and 100,000 deaths.[58] This is assuming stable atmospheric conditions, that the population had no warning, and that they had no protection from the agent (these same assumptions will be used throughout this article in describing WHO attack scenario estimates). In 2001, the Brentwood Postal Processing and Distribution Center in Washington, DC, had 2 envelopes pass through it that contained *B anthracis* spores. Spores leaked from the envelopes, became aerosolized through routine processing, and resulted in 4 deaths.[59,60] Despite widespread contamination of this 500,000-square foot facility and potential exposure of many workers, no other cases were reported.[60] One explanation for the lack of additional cases was the administration of antimicrobials to workers after exposure.

Botulism—The 7 toxins (types A through G) produced by *Clostridium botulinum* are the most toxic compounds known, with a toxic dose of 1 ng/kg (0.45 ng/lb) of body weight.[61] They are approximately 100,000 times as toxic as sarin.[61] In humans, the 3 naturally occurring forms of botulism are food-borne (involving preformed toxin), wound, and intestinal (infant) botulism[62]; inhalational botulism (often associ-

ated with laboratory exposure) and wound botulism are rare.[63] All forms of botulism result from the absorption of botulinum toxin into the bloodstream.[64] On aerosolization, the median lethal dose (LD_{50}) of *C botulinum* toxin is modified from 1 ng/kg to approximately 3 ng/kg (1.36 ng/lb) of body weight.[63] Because of this, a terrorist is more likely to disperse this agent through routes of ingestion rather than via aerosol. In humans, the onset of clinical signs varies with dose and may range from one to several days.[61] Clinical signs of botulism include symmetric descending flaccid paralysis that always begins in the bulbar musculature.[64] Patients have bulbar palsies with blurred vision, diplopia, ptosis, indistinct speech, difficulty swallowing, and progressive weakness from muscle paralysis leading to respiratory failure and death.[61,63] Because botulism is an intoxication, patients remain afebrile unless they acquire a secondary infection.[64] Paralysis can persist for weeks to months while affected individuals are receiving medical care. Treatment is supportive and requires early passive immunization with a trivalent equine antitoxin, which provides neutralizing antibodies to botulinum toxins A, B, and E and is only available from the CDC.[64] A heptavalent antitoxin containing antibodies against all 7 types of toxins is held by the US Army.[64] A pentavalent (ABCDE) botulinum toxoid vaccine is available through the CDC.[63,64] With proper medical management, including ventilatory assistance, deaths are likely to occur in < 5% of human cases.[61]

Dogs (and likely cats) with botulism develop ascending weakness from hind limbs to forelimbs, with hyporeflexia, hypotonia, weak vocalization, decreased gag reflex and jaw tone, slow pupillary reflexes, and eventually respiratory paralysis and death.[65] Recovery, if it occurs, is usually complete. In livestock, botulism occurs primarily when feed contaminated with preformed toxin is ingested. Sometimes this is a result of ingestion of feed contaminated with carrion (such as portions of dead carcasses of birds, rodents, or other animals on pasture that become incorporated into bales of hay) or the ingestion of poultry manure, spoiled silage or hay, or decaying vegetables.[66] Clinical signs in livestock may include muscle tremors, progressive weakness, incoordination, knuckling, ataxia, motor paralysis leading to recumbency, mydriasis, ptosis, and eventually death.

Plague—*Yersinia pestis*, the agent that causes plague, is found in natural enzootic foci throughout the world, with the exception of Australia and western Europe.[67,68] Within the United States, this agent is confined primarily to the western states; 90% or more of all human cases are reported in California, Colorado, Arizona, and New Mexico.[69] In humans, plague most often develops clinically as 1 of 3 forms: bubonic, septicemic, or pneumonic (primary or secondary) plague. It should be noted that the clinical aspects of plague compose a continuum and that illness, if untreated, will often progress from 1 form to the next (eg, bubonic to septicemic, then finally pneumonic). Most human disease is acquired from the bites of infected fleas that often result in regional lymphadenopathy (bubonic plague) and development of an ulcer or pustule at the site of inoculation. Common signs in patients with bubonic plague include local lymphadenopathy that is very painful, fever, chills, headache, and weakness.[70] Septicemic plague develops when *Y pestis* enters the bloodstream as a result of untreated bubonic plague (secondary septicemia) or in the absence of obvious lymph node swelling (primary septicemia).[67,70] Signs include disseminated intravascular coagulation, multiple organ failure, and adult respiratory distress syndrome.[71] Disseminated intravascular coagulation can lead to arteriolar thrombosis, hemorrhage in the skin, serosal surfaces or organs, and acral cyanosis and necrosis. The ensuing gangrene is believed to be the basis for the name, black death. Hematogenous spread of bacilli from bubonic or septicemic plague to the lungs results in secondary pneumonic plague. The resultant plague pneumonia, in humans and particularly cats, can then be spread via respiratory aerosol, initiating primary pneumonic plague in contacts and potentially resulting in an epidemic.[67,69] Primary pneumonic plague can also develop from the direct inhalation of aerosolized *Y pestis*, as might occur in a bioterrorism attack. Finally, transmission can also occur via direct handling of infective tissues.[67]

Aerosolization of *Y pestis* in a biological attack would pose a great threat to human health. In humans, primary pneumonic plague has a very rapid incubation period of 1 to 6 days (typically 2 to 4 days) and is usually fatal if not treated within the first 24 hours of illness.[70] Signs of primary pneumonic plague in humans include fever with cough and dyspnea; absence of buboes; watery, bloody, or purulent sputum; hemoptysis; nausea; vomiting; abdominal pain; and diarrhea.[70] In the United States, person-to-person spread of pneumonic plague has not been documented since 1924 to 1925.[67,69,72] Case-fatality rates for untreated bubonic plague range from 40% to 60%, and untreated septicemic and pneumonic disease are almost always fatal.[72]

Infection with *Y pestis* has been identified in numerous animal species, including > 200 species of rodents[67]; the highest death rates occur among rodents (especially prairie dogs [*Cynomys* spp], ground squirrels [*Spermophilus* spp], and antelope ground squirrels [*Ammospermophilus* spp]) and rabbits.[67,69] In other countries, camels and goats have developed *Y pestis*-associated illness,[73] but in the United States, livestock is not known to have developed illness.[69] Carnivores (dogs, ferrets, bears, coyotes, raccoons, and skunks) are fairly resistant to plague.[72] In naturally acquired plague, dogs will rarely have clinical signs (such as fever, lethargy, and possibly lymphadenopathy) and death is even more rare, if it occurs at all.[74] However, unlike dogs, cats readily develop plague[75] including secondary pneumonic plague and may directly infect humans through respiratory droplet transmission. Common clinical signs of naturally acquired plague in cats include lethargy, anorexia, fever, swollen lymph nodes, dyspnea, coughing, discharge from the mouth or nose, respiratory distress, septicemia, disseminated intravascular coagulation, and death.[75] From 1977 to 1998, there were 23 human cases of primary pneumonic

plague directly acquired from ill cats[76]; 6 (26.1%) of these were veterinarians or members of their staff.

The use of *Y pestis* as a biological weapon would most likely be in an aerosolized form and would drastically alter the natural epidemiologic features of this disease.[70] Via aerosol dispersal, the organism would infect humans as well as susceptible animals (primarily cats) that would pose a significant public health risk by serving as a source for continued transmission to humans. With the exception of cats, most domestic animals are fairly resistant to naturally occurring infection with *Y pestis*, and it is not known if dogs or livestock would develop pneumonic plague if they were to inhale large amounts of the agent. To the author's knowledge, there are no reports of primary pneumonic plague or experimental trials with aerosolized *Y pestis* in domestic animals. Findings that would suggest an intentional release of *Y pestis* would include detection of clusters of cases of pneumonic plague among humans or cats anywhere within the United States or isolated cases occurring outside the enzootic regions of the western United States. Enhanced awareness of bioterrorism among physicians was likely a contributing factor to the detection (in November 2002) of the first cases of human plague in New York City in more than 100 years.[77,78] A thorough history determined that the affected couple was most likely infected on their own property in New Mexico, where *Y pestis*-infected rodents and fleas had been found by the New Mexico Department of Health earlier in the year,[79] and not through a bioterrorist attack.

Biological attack scenario involving *Y pestis*

The WHO has estimated that under specific conditions, 50 kg of *Y pestis* released near a developed city of 5 million inhabitants would result in 150,000 illnesses and 36,000 deaths.[58]

Smallpox—There are approximately 12 known poxviruses (all included in 4 genera of the subfamily Chordopoxvirinae) that are zoonotic; infections with many of these viruses are rare. Smallpox has not been reported to be among them (**Appendix 2**). Indeed, smallpox and molluscum contagiosum are the only poxvirus infections not acquired directly or indirectly from animals.[80] An animal reservoir[88] for the smallpox virus has not been documented, and no cases of smallpox in animals have been reported. Experimental inoculation of animals with the smallpox virus has demonstrated development of little to no disease. Intradermal inoculation of 2 young camels with variola virus by Baxby et al[89] resulted in only minimal visible clinical effects; there was evidence of an inflamed swelling that disappeared within 1 week in both camels inoculated. Despite their attempts, those investigators were unable to isolate the virus from the swelling or blood samples.

Transmission of the variola virus that causes smallpox is most often by direct means, such as inhalation of droplets (ie, face-to-face contact) generated from the coughing or sneezing of an infected person, or via transfer of infectious substances (eg, scab material or pustular fluid) on the fingers after contact.[90,91] Indirect transmission is less common and can occur through contact with fomites (eg, blankets and sheets) contaminated by droplets, scabs, or pustular fluid. Aerosol transmission also may occur, but appears to be quite rare, especially over long distances.[90] In humans, smallpox has an incubation period of 7 to 17 days (typically 12 to 14 days)[91] at the end of which there is onset of fever, malaise, headache, backache, prostration, and possibly vomiting.[90] A maculopapular rash begins in the pharynx and mouth as the fever wanes and spreads to the face and remainder of the body. The rash progresses to vesicles, then pustules (described by some people as being hard as peas), with the formation of scabs by the eighth or ninth day after detection of the rash.[91] Compared with lesions associated with chickenpox, lesions of smallpox usually develop at the same rate and appear identical on a given region of the body.[91] Smallpox lesions are usually more dense over the face, arms, and legs (including palms and soles) than on the trunk, whereas chickenpox lesions are more dense over the trunk.[91] Patients are most infective in the first 7 to 10 days after the onset of the rash, but remain infective until scabs fall off. Little, if any, transmission occurs before the first day of visible rash.[92] By the time rash is detected, many patients are already restricted to bed because of the prodromal illness. Typical case-fatality rate with smallpox is 30% or higher in unvaccinated, untreated individuals.[91] Vaccination before exposure or within 2 to 3 days after exposure may provide near complete protection from development of disease; vaccination within 4 to 5 days after exposure may protect against death.[91]

In 2002, President Bush announced plans to vaccinate 500,000 health care workers (on a voluntary basis) in 2003, along with the mandatory vaccination of 500,000 Department of Defense soldiers and civilians deemed at highest risk should a biological attack involving smallpox occur.[93] The vaccine is based on the vaccinia virus; it is an effective vaccine of which the exact origins are unknown.[94] Vaccination against smallpox carries the risk of serious adverse effects (including death) among all who receive the vaccination, but these outcomes are highest in certain groups. Because of this, the CDC recommends that individuals who have any one of several preexisting medical conditions (eg, those persons who are organ transplant recipients; have received burns; or have eczema, shingles, human immunodeficiency virus infection, severe acne, or a heart condition), are pregnant or plan to become pregnant within 1 month of vaccination, or are < 18 years of age should not be vaccinated unless exposed to smallpox, nor should anyone who lives with them be vaccinated for fear of inadvertent inoculation.[95] Complications resulting from vaccination in the programs being conducted in military personnel and civilian health care and public health workers appear, for the most part, to be fewer than or similar to those associated with previous vaccination efforts. Between December 13, 2002, and May 28, 2003, 450,293 military personnel were vaccinated (70.5% of which were previously unvaccinated) against smallpox.[96] Within this population, the rates of generalized vaccinia (approx 80 cases/million persons vaccinated), inadvertent self inoculation (ie, transfer of vaccina virus from the vaccination site to other parts of the body such as face, eyes, nose, mouth, or genitalia via

hands contaminated after touching the vaccination site or dressings; approx 107 cases/million persons vaccinated), encephalitis (approx 2.2 cases/million persons vaccinated), and acute myopericarditis (approx 82 cases/million persons vaccinated) occurred within, or were less than, those of rates reported historically.[96] Only vaccinia transfer to a contact (approx 47 cases/million persons vaccinated) appeared to be higher than expected. Between January 24 and November 30, 2003, 38,908 civilian health care and public health workers were vaccinated.[97] The rate for self-inoculation (including transference to the eyes; approx 591 cases/million persons vaccinated) in this group was higher than previously recorded rates,[98,99] and the development of myocarditis or pericarditis (approx 565 cases/million persons vaccinated) was much higher than that observed in the military cohort that was vaccinated.[100] Overall, the vaccination of health care workers has been associated with at least 90 serious health events (ie, those requiring hospitalization, causing permanent disability, or resulting in severe illness or death) and at least 707 nonserious events.[97] Finally, there have been 3 deaths (2 health care workers[100] and 1 National Guardsman[96]) associated with myocardial infarctions that developed after vaccination, but there is no evidence to date that these deaths were causally linked to the smallpox vaccine.[101,102]

Some domestic animals (cows, water buffalo, pigs, and domestic rabbits) have become accidentally infected with vaccinia virus from vaccinated humans.[103,81] In 1964, a vaccinia outbreak involving 450 cows and 22 workers in an El Salvador dairy was traced back to 1 recently vaccinated worker.[104] However, the full potential for zoonotic transmission of the vaccinia virus and the public health implications are not known. Cantagalo virus is an apparent mutation of the vaccinia virus that was used during smallpox eradication efforts in Brazil decades ago; this virus appears to have become established in nature and is responsible for outbreaks in dairy cattle and their human contacts.[82] The animal reservoir for the Cantagalo virus is unknown. Given the present efforts to vaccinate persons in the United States against smallpox, the probability of domestic animals acquiring vaccinia infection and transmitting it to susceptible humans needs to be carefully considered. Whether animals that have been in contact with recently vaccinated humans could transmit vaccinia infection to persons with weakened immune systems has not been evaluated; because more vaccinations are planned, assessment of such risks seems prudent.

The recent multistate outbreak of monkeypox in the United States serves to highlight the fact that this naturally occurring agent may be employed by bioterrorists who may not be able to obtain variola virus, the official stocks of which are kept only in the United States and Russia. Prior to the US outbreak, monkeypox had occurred primarily in western and central African countries, where the animal reservoir is squirrels.[81] Squirrels can transmit the virus from one to another through direct or indirect contact (including via ingestion).[81] Transmission to humans from infected animals can occur through animal bites or from direct contact with animal blood, body fluids, or lesions. Person-to-person transmission of monkeypox occurs in the same manner as it does with smallpox (ie, via close person-to-person contact and transfer of respiratory droplets) but is rare, accounting for only 30% of observed cases in Africa.[81] Monkeypox virus is much less infective than variola virus.[105] Contaminated fomites may also serve as a source for infection. Assessments of outbreaks to date have revealed that transmission beyond the second generation (that is, beyond the first person-to-person transmission episode) of cases is rare.[80,83,106,107] The incubation period for monkeypox is approximately 12 days, and illness begins with fever, headache, muscle aches, backache, malaise, and lymphadenopathy.[105] A papular rash develops approximately 1 to 3 days after onset of fever, and lesions progress to scabs that eventually fall off.[105] In humans, the clinical features of monkeypox may closely resemble those of smallpox, except that lymphadenopathy is not observed in association with smallpox.[81] On the basis of historical evidence, vaccination of humans against smallpox appears to be approximately 85% effective in preventing monkeypox,[81,106,107] and individuals that have close contact with humans or animals that have monkeypox should receive the smallpox vaccine.[105] Case-fatality rates can range from 1% to 10%.[18,107]

Animals known to be susceptible to monkeypox include rodents, monkeys, and lagomorphs.[18] The susceptibility of many other animals, including dogs and cats, is unknown. Clinical features of the disease in animals would include a rash that is macular, papular, vesicular, or pustular in a localized or generalized distribution.[18] Other signs might include conjunctivitis, coryza, cough, anorexia, and lethargy. Among the prairie dogs involved in the US outbreak of monkeypox, clinical signs included fever, cough, conjunctivitis, lymphadenopathy, and nodular rash.[18] The CDC has released recommendations to veterinarians who might examine animals with monkeypox that include infection control precautions, personal protection measures, carcass disposal, and postexposure vaccination protocols.[18,108]

Tularemia—Tularemia is a naturally occurring disease found only in the northern hemisphere. It is caused by *Francisella tularensis*, which was first isolated in 1911 in an outbreak among rodents in Tulare County, California.[109] Natural reservoirs of the organism are small mammals, including rabbits, voles, water rats, muskrats, and ground squirrels.[109,110] *Francisella tularensis* may survive for a few weeks in soil, vegetation, and water[109,110] and for as long as 6 months in straw litter.[111] However, environmental survival of *F tularensis* after intentional release is likely to be minimal.[110] Humans acquire infection through the bites of arthropods (such as ticks and biting flies); handling of infected tissues or fluids; inhalation of aerosol; ingestion of contaminated food or water; or direct contact with contaminated food, water, or soil.[110] To cause illness in humans, 10 to 50 organisms are sufficient if inhaled or injected ID; however, a dose in the order of 10^8 organisms is required if the route of infection is via ingestion.[111,112] Aerosol exposure often occurs in situations in which people work with rodent-contaminated hay, thresh corn, or work in a laboratory.[113] In 2000, there was an outbreak of tularemia involving 15 humans in Martha's

Vineyard; the disease in 11 of these cases was classified as primary pneumonic.[114] The outbreak occurred as a result of aerosolization of *F tularensis* in animal excreta or rabbit carcasses during lawn mowing or the cutting of brush. Between 1990 and 2000, there were 1,368 cases of tularemia reported from 44 states in the United States.[115] Tularemia was removed from the list of notifiable diseases in 1994, but was reinstated in 2000 out of concern regarding its use in bioterrorism.[115]

In humans infected with *F tularensis*, there are 6 primary clinical presentations; the form of the disease that develops depends on the route of exposure and whether treatment is administered in an appropriate time period. These forms of tularemia are classified as glandular, ulceroglandular, oculoglandular, oropharyngeal, typhoidal, and pneumonic.[110,111] All forms can progress to pneumonia, sepsis, and death.[111] Typhoidal disease (ie, systemic infection with fever and signs associated with the gastrointestinal tract, but without cutaneous lesions or lymphadenopathy) results from exposure via inhalation, ingestion, or intradermal inoculation; of all the forms of tularemia, this has the highest case-fatality rate (approx 35%) if untreated.[112] The most likely route for dispersal of *F tularensis* in a bioterrorist attack would be via aerosol[110,112]; most affected individuals would develop the typhoidal form of disease (ie, systemic illness) and, less commonly, the pneumonic form.[110] In humans, tularemia is associated with a lower fatality rate than that of anthrax or plague.[110] The risk of animal-to-human or arthropod-to-human transmission following an intentional release of this agent is considered very low if basic precautions are taken.[110] Person-to-person transmission has never been documented. Mild inhalational tularemia in people may be mistaken for Q fever infection.[110]

Among livestock, sheep appear to be most often affected[111] and, along with pigs, generally develop the most clinical signs; however, calves are somewhat resistant.[116] In affected livestock, there may be evidence of tick infestation. The typical clinical signs of tularemia include fever, stiff gait, diarrhea, weight loss, and recumbency.[116] There are limited data reported regarding aerosol exposure in animals. Mortality rates in sheep may range from 20% to 50%.[116] Ticks are the most common vector for transmission of *F tularensis* to farm animals. Although dogs and cats can also acquire infection from ticks, ingestion of infected wild rabbits or rodents appears to be a common means of transmission in these species.[117] Dogs are generally resistant to infection but may develop anorexia, fever, listlessness, draining abscesses, and myalgia or may die suddenly.[117] Clinical signs are more likely to develop in cats than in dogs,[111] and felids may be more susceptible. Disease in cats has not been well described; features can be similar to those observed in dogs, but might also include lymphadenopathy and organ involvement.[117]

Biological attack scenario involving *F tularensis*

A hypothetical release of *F tularensis* in a city of 100,000, without intervention, could result in 82,500 illnesses and 6,188 deaths with a case-fatality rate of 7.5%; 95% of patients would be hospitalized.[57] Costs associated with the aftermath of this type of bioterrorist attack could near $5.5 billion. Previously, the WHO estimated that under specific conditions, an aerosol release of *F tularensis* near a city of 5 million inhabitants would result in 250,000 illnesses and 19,000 deaths.[58]

Viral hemorrhagic fever—Of all microbial and viral agents considered for bioterrorism, none are as feared by the public as those belonging to the **viral hemorrhagic fever (VHF)** group. Ebola virus, with its high mortality rate and the attention it has received in news media, movies, and novels, epitomizes the public's perception of VHF viruses. However, there are many other VHF viruses (**Appendix 3**). Viral hemorrhagic fever viruses are RNA viruses with lipid envelopes and are susceptible to detergents, low-pH environments, and bleach.[118] However, they are also stable in neutral-pH, protein-rich environments and have been isolated from blood after several weeks of storage.[118] In humans, early clinical signs of VHF infection include fever, malaise, prostration, conjunctivitis, bradycardia, tachypnea, hypotension, and cutaneous flushing or skin rash.[118,119] As the disease progresses, hemorrhagic diathesis can occur (with the development of petechiae, mucous membrane and conjunctival hemorrhage, hematuria, hematemesis, and melena).[119] Disseminated intravascular coagulation may develop, and abnormalities in circulation result in circulatory shock.[118.119] Not all viruses within this group have the same ability to cause the classic VHF syndrome, and the exact clinical picture varies among agents. Case-fatality rates range from < 1% for Rift Valley fever virus to almost 90% for Ebola virus.[119] In general, the incubation periods for humans infected with these viruses range from 2 to 21 days.[119]

With the exception of Ebola and Marburg viruses, the reservoirs for all VHF viruses are known and include rodents or arthropod vectors, and it is through contact with these that humans naturally acquire infection.[118] Person-to-person transmission is common among the VHF viruses, although the flaviviruses and Rift Valley fever virus are not transmitted in this manner.[119] Crimean-Congo hemorrhagic fever has considerable person-to-person transmission,[119,120] but does not replicate to high concentrations in cell culture and would therefore be less attractive to terrorists.[119] Viral hemorrhagic fever viruses are considered highly infectious and stable by fine-particle aerosols,[118,121,122] but experts have still not determined to what extent person-to-person aerosol transmission occurs in the natural disease epidemiology of VHFs.[123] With regard to the Ebola virus, person-to-person aerosol transmission does not appear to play much, if any, of a role in natural disease.[119,121,123] Continuous transmission between humans of VHF viruses does not occur. Because of this and the fact that most person-to-person transmission of VHF viruses occurs via direct contact with virus-contaminated body fluids,[119] it is unlikely that VHF infections resulting from a bioterrorism attack will spread like pneumonic plague.[122] Viral hemorrhagic fever virus transmission requires close contact.[123] Experimental aerosol trials of several of the VHF viruses have been conducted in nonhuman primates and have resulted in successful transmission and infection in these animals.[119] In guinea pigs, Lassa fever virus can

take as long as 55 minutes to lose half of its LD_{50} potential when aerosolized.[123] Although hantaviruses appear to be quite stable and are commonly transmitted via aerosol, few are capable of causing hemorrhagic lesions or hemorrhagic fever with renal syndrome. Hantaviruses are classified as Category C agents, likely because they have almost no person-to-person transmission potential, are difficult to isolate and grow, and may lose infectivity in cell culture.[120] Compared with filoviruses and arenaviruses, hantaviruses are not useful as potential bioterrorism agents.[120]

In nature, arenaviruses are transmitted to humans via inhalation of rodent feces or urine contaminated with virus, ingestion of rodent-contaminated food, or direct contact with rodent excreta.[119] The arenaviruses appear more stable in the environment and in aerosolized forms than the other VHF agents[122] and can be transmitted person-to-person more readily.[119,124]

In domestic animals, primarily livestock, the clinical signs of Crimean-Congo hemorrhagic fever[125] may be mild and those of Rift Valley fever may be severe.[126] For all other VHF viruses, natural infection in domestic animals appears to result in no apparent illness. Rift Valley fever virus is restricted to the African continent and spreads between animals principally via mosquitoes.[127,128] This virus can infect many species of animals, including dogs and cats, and could be transmitted via arthropods that are found in the United States.[122,129] Disease is most severe in young animals. In ruminants, clinical signs of Rift Valley fever include biphasic fever, listlessness, depressed appetite, weight loss, abortion (which sometimes is the only clinical sign), and death.[130] Diarrhea, signs of abdominal pain, regurgitation, and necrotic hepatitis may also be observed. In epidemics of Rift Valley fever, sheep and cattle are the most important species and both may have abortion rates of 100% after infection; these species also have notable mortality rates after infection (as high as 90% in newborn lambs and 20% in pregnant ewes).[126] Dogs and cats < 3 weeks old that are infected with Rift Valley fever virus may have no signs of clinical illness or may develop diffuse petechiae, meningitis, myocarditis, and hepatic necrosis and die.[131] Older dogs and cats with infection appear to develop little disease. Nevertheless, the use of Rift Valley fever virus as a bioterrorism agent could serve a dual purpose, potentially infecting humans and livestock at the same time; because the virus is classified as a List A disease by the Office International des Épizooties, its discovery within US borders would likely result in severe economic repercussions from loss in trade.[127]

Vaccines for humans against Rift Valley fever[132,133] and Junin[119,124] are available as investigational new drugs, and a vaccine against Kyasanur Forest disease is in the early stages of development.[134] A vaccine for humans against Ebola is not available,[121,135] but development recently of an accelerated vaccine that confers protection in nonhuman primates suggests that a vaccine for human use is under investigation.[84] Laboratory testing for VHF agents is more limited than for other Category A and B agents, requiring specialized facilities and safety equipment. At a minimum, **biosafety level (BSL)**-3 facilities are needed for handling clinical specimens, and high-containment BSL-4 facilities are required for viral isolation.[119] There are only 2 BSL-4 facilities in the country with diagnostic capabilities: 1 is located at the CDC and the other at the **US Army Medical Research Institute of Infectious Diseases (USAMRIID)**.[119] As clinical microbiology and public health laboratories are not equipped to perform diagnostic testing for VHF viruses, all specimens must be sent to the CDC or USAMRIID.[121] This limited laboratory capability is a concern and a factor to consider in the development of bioterrorism response plans.

Conclusion

Of the 6 groups of agents in Category A (as designated by the CDC), all pose a tremendous risk to humans and 5 pose a risk to animals. The zoonotic potential of these agents should not be overlooked, nor should the role of veterinarians in detection of these diseases in either animals or humans who are in contact with them. The threat of bioterrorism is not likely to disappear in the near future. Veterinarians should be able to recognize the rudimentary clinical signs of diseases caused by Category A agents, amongst others; they should also understand that many people consider veterinarians to have expert knowledge regarding these agents. The time to learn about the Category A, B, and C agents is before a bioterrorist attack, not after.

References

1. Barton R. The application of the UNSCOM experience to international biological arms control. *Crit Rev Microbiol* 1998;24:219–233.
2. Mangold T, Goldberg J. *Plague wars: the terrifying reality of biological warfare*. New York: St Martin's Press, 1999.
3. Miller J, Engelberg S, Broad W. *Germs: biological weapons and America's secret war*. New York: Simon & Schuster, 2001;15–33.
4. Zilinskas RA. Iraq's biological weapons: the past as future? *JAMA* 1997;278:418–424.
5. World Health Organization. Severe acute respiratory syndrome. *Wkly Epidemiol Rec* 2003;78:81–83.
6. World Health Organization. Cumulative number of reported probable cases of SARS. Available at: www.who.int/csr/sars/country/2003_07_11/en/. Accessed Jul 31, 2003.
7. Centers for Disease Control and Prevention. Update: Severe acute respiratory syndrome—worldwide and United States, 2003. *MMWR Morb Mortal Wkly Rep* 2003;52:664–665.
8. Drosten C, Gunther S, Preiser W, et al. Identification of a novel coronavirus in patients with severe acute respiratory syndrome. *N Engl J Med* 2003;348:1967–1976.
9. Ksiazek TG, Erdman D, Goldsmith C, et al. A novel coronavirus associated with severe acute respiratory syndrome. *N Engl J Med* 2003;348:1953–1966.
10. Rota PA, Oberste MS, Monroe SS, et al. Characterization of a novel coronavirus associated with severe acute respiratory syndrome. *Science* 2003;300:1394–1398.
11. Breiman RF, Evans MR, Preiser W, et al. Role of China in the quest to define and control severe acute respiratory syndrome. *Emerg Infect Dis* 2003;9:1037–1041.
12. Guan Y, Zheng BJ, He YQ, et al. Isolation and characterization of viruses related to the SARS coronavirus from animals in southern China. *Science* 2003;302:276–278.
13. World Health Organization. Consensus document on the epidemiology of severe acute respiratory syndrome (SARS). Available at: www.who.int/csr/sars/en/index.html. Accessed Nov 20, 2003.
14. Centers for Disease Control and Prevention. Prevalence of IgG antibody to SARS-associated coronavirus in animal traders—Guangdong Province, China, 2003. *MMWR Morb Mortal Wkly Rep* 2003;52:986–987.
15. Martina BEE, Haagmans BL, Kuiken T, et al. SARS virus infection of cats and ferrets. *Nature* 2003;425:915.
16. Centers for Disease Control and Prevention. Multistate out-

break of monkeypox—Illinois, Indiana, and Wisconsin, 2003. *MMWR Morb Mortal Wkly Rep* 2003;52:537–540.

17. Centers for Disease Control and Prevention. Update: multistate outbreak of monkeypox—Illinois, Indiana, Kansas, Missouri, Ohio, and Wisconsin, 2003. *MMWR Morb Mortal Wkly Rep* 2003;52:642–646.

18. Centers for Disease Control and Prevention. Monkeypox infections in animals: updated interim guidance for veterinarians. Available at: www.cdc.gov/ncidod/monkeypox/index.htm. Accessed Dec 10, 2003.

19. Gage KL, Montenieri JA. The role of predators in the ecology, epidemiology, and surveillance of plague in the United States, in *Proceedings*. 16th Vertebr Pest Conf 1994;200–206.

20. Clark RK, Jessup DA, Hird DW, et al. Serologic survey of California wild hogs for antibodies against selected zoonotic disease agents. *J Am Vet Med Assoc* 1983;183:1248–1251.

21. Warner RD, Marsh WW. Rocky Mountain spotted fever. *J Am Vet Med Assoc* 2002;221:1413–1417.

22. Gage KL. Plague surveillance. In: *Plague manual: epidemiology, distribution, surveillance and control*. Geneva: World Health Organization, 1999;135–161.

23. van der Schalie WH, Gardner HS Jr, Bantle JA, et al. Animals as sentinels of human health hazards of environmental chemicals. *Environ Health Perspect* 1999;107:309–315.

24. Huxsoll DL, Patrick WC III, Parrott CD. Veterinary services in biological disasters. *J Am Vet Med Assoc* 1987;190:714–722.

25. Todd FA. Biological warfare against our livestock. *North Am Vet* 1952;33:689–693.

26. Federal Bureau of Investigation. *Terrorism in the United States, 1999*. Washington, DC: Federal Bureau of Investigation, Counterterrorism, Threat Assessment and Warning Unit, Counterterrorism Division, 1999.

27. Lesser IO, Hoffman B, Arquilla J, et al. *Countering the new terrorism*. Washington, DC: RAND, 1999.

28. September 11, 2001 Victims. September 11, 2001 victims home page. Available at: www.september11victims.com/september11victims/. Accessed May 6, 2003.

29. Jernigan DB, Raghunathan PL, Bell BP, et al. Investigation of bioterrorism-related anthrax, United States, 2001: epidemiologic findings. *Emerg Infect Dis* 2002;8:1019–1028.

30. Török TJ, Tauxe RV, Wise RP, et al. A large community outbreak of salmonellosis caused by intentional contamination of restaurant salad bars. *JAMA* 1997;278:389–395.

31. United States Department of State. *Patterns of global terrorism—2001*. Washington, DC: US Department of State, 2002.

32. Carus WS. Center for Counterproliferation Research, National Defense University. Working paper: bioterrorism and biocrimes. The illicit use of biological agents since 1900. Available at: www.ndu.edu/centercounter/Full_Doc.pdf. Accessed Jun 1, 2002.

33. Stern J. *The ultimate terrorists*. London: Harvard University Press, 1999.

34. Rotz LD, Khan AS, Lillibridge SR, et al. Public health assessment of potential biological terrorism agents. *Emerg Infect Dis* 2002;8:225–230.

35. Bell JH. Anthrax and the wool trade. *Am J Public Health* 2002;92:754–757.

36. Brachman PS. Inhalation anthrax. *Ann N Y Acad Sci* 1980;353:83–93.

37. Jernigan JA, Stephens DS, Ashford DA, et al. Bioterrorism-related inhalational anthrax: the first 10 cases reported in the United States. *Emerg Infect Dis* 2001;7:933–944.

38. Inglesby TV, O'Toole T, Henderson DA, et al. Anthrax as a biological weapon, 2002. *JAMA* 1999;281:2236–2252.

39. Friedlander AM. Anthrax. In: Zajtchuk R, Bellamy RF, Sidell FR, et al, eds. *Textbook of military medicine, medical aspects of chemical and biological warfare. Part I*. Washington, DC: Office of The Surgeon General, Department of the Army, 1997;467–478.

40. Turnbull PCB. *Guidelines for the surveillance and control of anthrax in humans and animals*. 3rd ed. Geneva: World Health Organization, 2002.

41. Centers for Disease Control and Prevention. Human anthrax associated with an epizootic among livestock—North Dakota, 2000. *MMWR Morb Mortal Wkly Rep* 2001;50:677–680.

42. Swartz MN. Recognition and management of anthrax—an update. *N Engl J Med* 2001;345:1621–1626.

43. Cieslak TJ, Eitzen EM Jr. Clinical and epidemiologic principles of anthrax. *Emerg Infect Dis* 1999;5:552–555.

44. Suffin SC, Carnes WH, Kaufmann AF. Inhalation anthrax in a home craftsman. *Hum Pathol* 1978;9:594–597.

45. Friedlander AM, Welkos SL, Pitt MLM, et al. Postexposure prophylaxis against experimental inhalation anthrax. *J Infect Dis* 1993;167:1239–1242.

46. Glassman HN. Industrial inhalational anthrax. *Bacteriol Rev* 1966;30:657–659.

47. Jefferds DM, Laserson K, Fry AM, et al. Adherence to antimicrobial inhalational anthrax prophylaxis among postal workers, Washington, D.C., 2001. *Emerg Infect Dis* 2002;8:1138–1144.

48. Centers for Disease Control and Prevention. Additional options for preventive treatment for persons exposed to inhalational anthrax. *MMWR Morb Mortal Wkly Rep* 2003;50:1142, 1151.

49. Peters CJ, Hartley DM. Anthrax inhalation and lethal human infection. *Lancet* 2002;359:710–711.

50. Williams AA, Parashar UD, Stoica A, et al. Bioterrorism-related anthrax surveillance, Connecticut, September–December, 2001. *Emerg Infect Dis* 2002;8:1078–1082.

51. Whitford HW, Hugh-Jones ME. Anthrax. In: Beran GW, Steele JH, eds. *Handbook of zoonoses, section A: bacterial, rickettsial, chlamydial, and mycotic zoonoses*. 2nd ed. Boca Raton, Fla: CRC Press Inc, 1994;61–82.

52. Radostits OM, Gay CC, Blood DC, et al. *Veterinary medicine: a textbook of the diseases of cattle, sheep, pigs, goats and horses*. 9th ed. London: Harcourt Publishers Ltd, 2000;747–751.

53. Greene CE, Prescott JF. Streptococcal and other gram-positive bacterial infections. In: Green CE, eds. *Infectious diseases of the dog and cat*. 2nd ed. Philadelphia: WB Saunders Co, 1998;205–214.

54. McGee ED, Fritz DL, Ezzell JW, et al. Anthrax in a dog. *Vet Pathol* 1994;31:471–473.

55. Gleiser CA, Gochenour WS, Ward MK. Pulmonary lesions in dogs and pigs exposed to a cloud of anthrax spores. *J Comp Pathol* 1968;78:445–449.

56. Bales ME, Dannenberg AL, Brachman PS, et al. Epidemiologic response to anthrax outbreaks: field investigations, 1950–2001. *Emerg Infect Dis* 2002;8:1163–1174.

57. Kaufmann AF, Meltzer MI, Schmid GP. The economic impact of a bioterrorist attack: are prevention and postattack intervention programs justifiable? *Emerg Infect Dis* 1997;3:83–94.

58. World Health Organization. *Health aspects of chemical and biological weapons*. Geneva: World Health Organization, 1970;1–132.

59. Centers for Disease Control and Prevention. Evaluation of *Bacillus anthracis* contamination inside the Brentwood mail processing and distribution center—District of Columbia, October, 2001. *MMWR Morb Mortal Wkly Rep* 2001;50:1129–1133.

60. Dewan PK, Fry AM, Laserson K, et al. Inhalational anthrax outbreak among postal workers, Washington, D.C., 2001. *Emerg Infect Dis* 2003;8:1066–1072.

61. Franz DR, Jahrling PB, Friedlander AM, et al. Clinical recognition and management of patients exposed to biological warfare agents. *JAMA* 1997;278:399–411.

62. American Public Health Association. In: Chin J, eds. *Control of communicable disease manual*. 17th ed. Washington, DC: American Public Health Association, 2000;70–74.

63. Middlebrook JL, Franz DR. Botulinum toxins. In: Zajtchuk R, Bellamy RF, Sidell FR, et al, eds. *Textbook of military medicine, medical aspects of chemical and biological warfare. Part I*. Washington, DC: Office of The Surgeon General, Department of the Army, 1997;643–654.

64. Arnon SS, Schechter R, Inglesby TV, et al. Botulinum toxin as a biological weapon. *JAMA* 2001;285:1059–1070.

65. Barsanti JA. Botulism. In: Greene CE, eds. *Infectious diseases of the dog and cat*. 2nd ed. Philadelphia: WB Saunders Co, 1998;263–267.

66. Radostits OM, Gay CC, Blood DC, et al. *Veterinary medicine: a textbook of the diseases of cattle, sheep, pigs, goats and horses*. 9th ed. London: Harcourt Publishers Ltd, 2000;757–760.

67. Poland JD, Quan TJ, Barnes AM. Plague. In: Beran GW, Steele JH, eds. *Handbook of zoonoses, section A: bacterial, rickettsial, chlamydial, and mycotic zoonoses*. 2nd ed. Boca Raton, Fla: CRC Press Inc, 1994;93–112.

68. Tikhomirov E. Epidemiology and distribution of plague. *Plague manual: epidemiology, distribution, surveillance and control*. Geneva: World Health Organization, 1999;11–42.

69. Orloski KA, Lathrop SL. Plague: a veterinary perspective. *J Am Vet Med Assoc* 2003;222:444–448.

70. Inglesby TV, Dennis DT, Henderson DA, et al. Plague as a biological weapon. *JAMA* 2000;283:2281–2290.

71. Poland JD, Dennis DT. Diagnosis and clinical manifestations. In: *Plague manual: epidemiology, distribution, surveillance and control*. Geneva: World Health Organization, 1999;43–53.

72. Perry RD, Fetherston JD. *Yersinia pestis*—etiologic agent of plague. *Clin Microbiol Rev* 1997;10:35–66.

73. Christie AB, Chen TH, Elsberg SS. Plague in camels and goats: their role in human epidemics. *J Infect Dis* 1980;141: 724–726.

74. Orloski KA, Eidson M. *Yersinia pestis* infection in three dogs. *J Am Vet Med Assoc* 1995;207:316–318.

75. Eidson M, Thilsted JP, Rollag OJ. Clinical, clinicopathologic, and pathologic features of plague in cats: 119 cases (1977–1988). *J Am Vet Med Assoc* 1991;199:1191–1197.

76. Gage KL, Dennis DT, Orloski KA, et al. Cases of cat-associated human plague in the western US, 1977–1998. *Clin Infect Dis* 2000;20:893–900.

77. Ackelsberg J, Layton M. New York County Medical Society. 2002 Health Alert #36. Two cases of travel-associated bubonic plague in New York City. Available at: www.nycms.org/article_view.php3?view=529&part=1. Accessed Dec 12, 2003.

78. CNN. Bubonic plague suspected in NYC visitors. Available at: www.cnn.com/2002/US/Northeast/11/06/ny.plague/. Accessed Dec 12, 2003.

79. Centers for Disease Control and Prevention. Imported plague—New York City, 2002. *MMWR Morb Mortal Wkly Rep* 2003;52:725–728.

80. Baxby D, Bennett M. Poxvirus zoonoses. *J Med Microbiol* 1997;46:17–20, 28–33.

81. Fenner F. Poxviral Zoonoses. In: Beran GW, Steele JH, eds. *Handbook of zoonoses, section B: viral zoonoses*. 2nd ed. Boca Raton, Fla: CRC Press Inc, 1994;485–503.

82. Damaso CR, Esposito JJ, Condit RC, et al. An emergent poxvirus from humans and cattle in Rio de Janeiro State: Cantagalo virus may derive from Brazilian smallpox vaccine. *Virology* 2000;277:439–449.

83. Pattyn SR. Monkeypoxvirus infections. *Rev Sci Tech* 2000; 19:92–97.

84. Sullivan NJ, Geisbert TW, Geisbert JB, et al. Accelerated vaccination for Ebola virus haemorrhagic fever in non-human primates. *Nature* 2003;424:681–684.

85. Fenner F. Poxviruses. In: Fields B, Knipe D, Howley P, et al, eds. *Fields virology*. 3rd ed. Philadelphia: Lippincott-Raven Publishers, 1996;2673–2702.

86. Kolhapure RM, Deolanker RP, Tupe CD, et al. Investigation of buffalopox outbreaks in Maharashtra State during 1992–1996. *Indian J Med Res* 1997;106:441–446.

87. Sawhney AN, Singh AK, Malik BS. Goat-pox: an anthropozoonosis. *Indian J Med Res* 1972;60:683–684.

88. Fenner F, Henderson DA, Arita I, et al. *Smallpox and its eradication*. Geneva: World Health Organization, 1988;479–480.

89. Baxby D, Ramyar H, Hessami M, et al. Response of camels to intradermal inoculation with smallpox and camelpox viruses. *Infect Immun* 1975;11:617–621.

90. Fenner F, Henderson DA, Arita I, et al. *Smallpox and its eradication*. Geneva: World Health Organization, 1988;191–194.

91. Henderson DA, Inglesby TV, Bartlett JG, et al. Smallpox as a biological weapon. *JAMA* 1999;281:2127–2137.

92. Fenner F, Henderson DA, Arita I, et al. *Smallpox and its eradication*. Geneva: World Health Organization, 1988;189.

93. Pilch R. Monterey Institute of International Studies. Smallpox: threat, vaccine, and U.S. policy, Part 1 of 6. Available at: cns.miis.edu/pubs/week/030106.htm. Accessed Mar 5, 2003.

94. Fenner F, Henderson DA, Arita I, et al. *Smallpox and its eradication*. Geneva: World Health Organization, 1988;278.

95. Centers for Disease Control and Prevention. People who should not get the smallpox vaccine. Smallpox fact sheet. Available at: www.bt.cdc.gov/agent/smallpox/vaccination/contraindications-public.asp. Accessed Dec 12, 2003.

96. Grabenstein JD, Winkenwerder W Jr. US military smallpox vaccination program experience. *JAMA* 2003;289:3278–3282.

97. Centers for Disease Control and Prevention. Smallpox vaccination program report status and adverse events. Available at: www.cdc.gov/od/oc/media/smpxrprt.htm. Accessed Dec 10, 2003.

98. Lane JM, Ruben FL, Neff JM, et al. Complications of smallpox vaccination, 1968: national surveillance in the United States. *N Engl J Med* 1969;281:1201–1208.

99. Lane JM, Ruben FL, Neff JM, et al. Complications of smallpox vaccination, 1968: results of ten statewide surveys. *J Infect Dis* 1970;122:303–309.

100. Centers for Disease Control and Prevention. Update: cardiac-related events during the civilian smallpox vaccination program—United States, 2003. *MMWR Morb Mortal Wkly Rep* 2003;52:492–496.

101. Centers for Disease Control and Prevention. Cardiac deaths after a mass smallpox vaccination campaign—New York City, 1947. *MMWR Morb Mortal Wkly Rep* 2003;52:933–936.

102. Ault A. US smallpox vaccine programme stalls as volunteers balk. *Lancet* 2003;361:1626.

103. Dekking F. Cowpox and vaccinia. In: van der Hoeden J, ed. *Zoonoses*. Amsterdam: Elsevier Pub Co, 1964;411–418.

104. Lum GS, Soriano F, Trejos A, et al. Vaccinia epidemic and epizootic in El Salvador. *Am J Trop Med Hyg* 1967;16:332–338.

105. Centers for Disease Control and Prevention. Basic information about monkeypox. Available at: www.cdc.gov/ncidod/monkeypox/factsheet.htm. Accessed Dec 10, 2003.

106. Fine PE, Jezek Z, Grab B, et al. The transmission potential of monkeypox virus in human populations. *Int J Epidemiol* 1988;17:643–650.

107. Hutin YJ, Williams RJ, Malfait P, et al. Outbreak of human monkeypox, Democratic Republic of Congo, 1996–1997. *Emerg Infect Dis* 2001;7:434–438.

108. Centers for Disease Control and Prevention. Smallpox vaccination and monkeypox. Available at: www.cdc.gov/ncidod/monkeypox/smallpoxvaccine_mpox.htm. Accessed Dec 10, 2003.

109. Ellis J, Oyston PCF, Green M, et al. Tularemia. *Clin Microbiol Rev* 2002;15:631–646.

110. Dennis DT, Inglesby TV, Henderson DA, et al. Tularemia as a biological weapon. *JAMA* 1999;285:2763–2773.

111. Feldman KA. Tularemia. *J Am Vet Med Assoc* 2003;222: 725–730.

112. United States Army Medical Research Institute of Infectious Diseases. Tularemia. In: Kortepeter M, ed. *Medical management of biological casualties handbook: the blue book*. 4th ed. Fort Detrick, Md: US Army Medical Research Institute of Infectious Diseases, 2001;69–77.

113. Pearson A. Tularemia. In: Palmer SR, Soulsby L, Simpson DIH, eds. *Zoonoses*. New York: Oxford University Press, 1998; 267–279.

114. Feldman KA, Enscore RE, Lathrop SL, et al. An outbreak of primary pneumonic tularemia on Martha's Vineyard. *N Engl J Med* 2001;345:1601–1606.

115. Centers for Disease Control and Prevention. Tularemia—United States, 1990–2000. *MMWR Morb Mortal Wkly Rep* 2002;51:181–184.

116. Radostits OM, Gay CC, Blood DC, et al. *Veterinary medicine: a textbook of the diseases of cattle, sheep, pigs, goats and horses*. 9th ed. London: Harcourt Publishers Ltd, 2000;857–858.

117. Kaufmann AF. Tularemia. In: Greene CE, eds. *Infectious diseases of the dog and cat*. 2nd ed. Philadelphia: WB Saunders Co, 1998;300–302.

118. Jahrling PB. Viral hemorrhagic fevers. In: Zajtchuk R, Bellamy RF, Sidell FR, et al, eds. *Textbook of military medicine: medical aspects of chemical and biological warfare. Part I*. Washington, DC: Office of The Surgeon General, Department of the Army, 1997;591–602.

119. Borio L, Inglesby T, Peters CJ, et al. Hemorrhagic fever viruses as biological weapons. *JAMA* 2002;287:2391–2405.

120. Clement JP. Hantavirus. *Antiviral Res* 2003;57:121–127.

121. Bray M. Defense against filoviruses used as biological weapons. *Antiviral Res* 2003;57:53–60.

122. Peters CJ. Are hemorrhagic fever viruses practical agents for

biological terrorism? In: Scheld WM, Craig WA, Hughes JM, eds. *Emerging infections 4*. Washington, DC: American Society for Microbiology, 2000;201–209.

123. Peters CJ, Jahrling PB, Khan AS. Patients infected with high-hazard viruses: scientific basis for infection control. *Arch Virol Suppl* 1996;11:141–168.

124. Charrel RN, Lamballerie X. Arenaviruses other than Lassa virus. *Antiviral Res* 2003;57:89–100.

125. Swanepoel R. Crimean-congo hemorrhagic fever. In: Beran GW, Steele JH, eds. *Handbook of zoonoses, section B: viral zoonoses*. 2nd ed. Boca Raton, Fla: CRC Press Inc, 1994;149–161.

126. Peters CJ, Linthicum KJ. Rift Valley fever. In: Beran GW, Steele JH, eds. *Handbook of zoonoses, section B: viral zoonoses*. 2nd ed. Boca Raton, Fla: CRC Press Inc, 1994;125–138.

127. Office International des Epizooties. Rift Valley fever. Available at: www.oie.int/eng/maladies/fiches/a_A080.htm. Accessed Mar 27, 2003.

128. Radostits OM, Gay CC, Blood DC, et al. *Veterinary medicine: a textbook of the diseases of cattle, sheep, pigs, goats and horses*. 9th ed. London: Harcourt Publishers Ltd, 2000;1042–1044.

129. Gargan TP II, Clark GG, Dohm DJ, et al. Vector potential of selected North American mosquito species for Rift Valley fever virus. *Am J Trop Med Hyg* 1988;38:440–446.

130. Barnhard BJH. Rift Valley fever. In: Aiello SE, Mays A, eds. *The Merck veterinary manual*. 8th ed. Whitehouse Station, NJ: Merck & Co Inc, 1998;541–542.

131. Greene CE, Baldwin CA. Arboviral infections. In: Greene CE, ed. *Infectious diseases of the dog and cat*. 2nd ed. Philadelphia: WB Saunders Co, 1998;131–132.

132. Niklasson B, Peters CJ, Bengtsson E, et al. Rift Valley fever virus vaccine trial: study of neutralizing antibody response in humans. *Vaccine* 1985;3:123–127.

133. Pittman PR, Liu CT, Cannon TL, et al. Immunogenicity of an inactivated Rift Valley fever vaccine in humans. *Vaccine* 1999;18:181–189.

134. Dandawate DN, Desai GB, Achar TR, et al. Field evaluation of formalin inactivated Kyasanur forest disease virus tissue culture vaccine in three districts of Karnataka state. *Indian J Med Res* 1994;99:152–158.

135. Geisbert TW, Pushko P, Anderson K, et al. Evaluation in nonhuman primates of vaccines against Ebola virus. *Emerg Infect Dis* 2002;8:503–507.

Appendix 1

Categorization of bioterrorism agents of public health importance (as designated by the Centers for Disease Control and Prevention)

Agent	Disease
Category A	
Bacillus anthracis	Anthrax
Clostridium botulinum (toxin)	Botulism
Yersinia pestis	Plague
Variola major	Smallpox
Francisella tularensis	Tularemia
Filoviruses and Arenaviruses (eg, Ebola and Lassa viruses)	Viral hemorrhagic fever
Category B	
Brucella spp	Brucellosis
Burkholderia mallei	Glanders
Burkholderia pseudomallei	Melioidosis
Chlamydophila psittaci	Psittacosis
Coxiella burnetii	Q fever
Rickettsia prowazekii	Epidemic typhus fever
Alphaviruses (western, eastern, and Venezuelan equine encephalitis viruses)	Encephalitis
Toxins (eg, ricin and staphylococcal enterotoxin B)	Toxic syndromes
Food safety threats (eg, *Salmonella* spp and *Escherichia coli* 0157:H7)	
Water safety threats (eg, *Cryptosporidium* spp and *Vibrio cholerae*)	
Category C	
Emerging threats (eg, Nipah virus and Hantavirus)	

Appendix 2

Zoonotic poxviruses and their animal reservoirs[80-87]

Genus	Virus	Reservoir[81,82]
Orthopoxvirus		
	Cowpox virus	Rodents
	Monkeypox virus	Squirrels
	Vaccinia virus	Unknown
	Buffalopox virus*	Buffaloes
	Cantagalo virus†	Unknown
Parapoxvirus		
	Bovine papular stomatitis virus	Cattle
	Pseudocowpox virus	Dairy cattle
	Orf virus	Sheep
	Sealpox virus	Seals
Capripoxvirus		
	Goatpox virus‡	Goats
Yatapoxvirus		
	Tanapox virus	Rodents
	Yaba poxvirus	Monkeys

*Considered by many to be a subspecies of vaccinia virus. †Newly described.[82] ‡Limited evidence.

Appendix 3

Viral hemorrhagic fever viruses and their potential for bioterrorism use

Family	Genus	Virus	Potential for use
Arenaviridae			
	Arenavirus	Lassa, New World Arenaviridae*	High
Bunyaviridae			
	Nairovirus	Crimean-Congo hemorrhagic fever	Low
	Phelbovirus	Rift Valley fever	High
	Hantavirus	Viruses that cause hemorrhagic fever with renal syndrome	Low
Filoviridae			
	Filovirus	Ebola	High
		Marburg	High
Flaviviridae			
	Flavivirus	Dengue	Low
		Kyasanur Forest disease	High
		Omsk hemorrhagic fever	High
		Yellow fever	High

*This includes many New World viruses such as Guanarito, Junin, Machupo, and Sabia, a new arenavirus discovered in California in 1999 and 2000.