

Bioterrorism and Emerging Infections Education

ANTHRAX

Anthrax is a zoonotic infection with a long association with human history. It is likely that the fifth and sixth plagues described in Exodus may have been anthrax in domestic animals and humans. In addition, during the 16th and 18th centuries in Europe, anthrax was an economically important agricultural disease. Inhalational anthrax was first described during the latter half of the 19th century. The disease was discovered among woolsorters in England, by John Bell in the late 15th century, caused by the generation of anthrax spore aerosols during the processing of goat wool.

Robert Koch established the microbial origin of anthrax in 1876 and discovered the sporulation processes and spore stage in *Bacillus anthracis*. He also developed the first nutrient media and cultivation techniques for *Bacillus anthracis*. Anthrax was also the first disease for which Louis Pasteur, in 1881, developed an effective live bacterial vaccine.

Anthrax occurs in domestic, wild and exotic animals, including goats, sheep, cattle, hippos, elephants, lions, zebras, and camels. Humans usually become infected via contact with infected animals or contaminated articles and animal products. Anthrax predominantly presents as a cutaneous infection, but may occur in gastrointestinal and inhalational form.

History of development as a weapon

Anthrax has been the focus of much attention as a potential biological weapon for at least eight decades. An early attempt to use anthrax spores as a weapon (against horses) occurred during World War I. Before and during World War II, the Japanese military experimented with anthrax, and was able to develop the first relatively sophisticated anthrax biological weapons. Since WWII, many countries, including Russia, Great Britain, Canada, the United States and Iraq, have shown a significant interest in the possible use of anthrax as an instrument of war.

Although many infectious agents can be used to develop biological weapons, anthrax has been identified historically as uniquely suited to this use for many reasons, including:

- **Relative ease of weapon production:** Both the U.S. and the former Soviet Union were capable of producing tons of dry formulation with a high concentration of spores per gram. Individual or sub-state terrorists could produce gram quantities, such as that seen in letters sent to the Hart Senate building in 2001, with relatively very simple equipment -- if they understand what they are doing.
- **Long shelf life:** The liquid form of anthrax weapon can be stored at 0°C for approximately one year. The dry form has a much longer shelf life; no decay was observed even after five years in storage. Spores can remain viable for many years, despite significant changes in light and temperature.

- **Effective dispersion:** As military weapons dispersion can be achieved by either explosion or high-speed/high-pressure air streams. Because anthrax spores have a durable outer shell, they are more likely to survive such dissemination than other biological agents. Terrorist use of anthrax formulations, in very small quantities, can be done much more simply, especially in confined spaces.
- **Persistence:** Anthrax spores are persistent and environmentally stable. The area of contamination from an aerosolized weapon would be very large and could remain contaminated for months or even years, at least in areas not exposed to sun and weather. During the 1979 accidental anthrax release in Sverdlosk the contaminated area extended 50 kilometers downwind from the production facility. Because of the agent's persistence in the environment, new cases of infection may occur in the forms of secondary aerosols created when the settled agent is disturbed. Although the threat of secondary aerosols is not fully understood, degree of re-aerosolization is dependent on both concentration of spores deposited and quality of formulation.
- **High mortality rate:** Systemic anthrax infection from anthrax spore inhalation causes septic shock with a mortality rate approaching 100%. Death occurs within a few days after the onset of symptoms.
- **Lack of effective treatment:** Once symptoms of inhalational anthrax appear, treatment is much less effective and the case-fatality rate remains very high. Historically, one might have expected to save only approximately 15% of patients presenting with inhalational anthrax. Modern medical care and clinician awareness following the first case in October 2001 resulted in saving 6 of 11 patients diagnosed with inhalational anthrax.

An inadvertent demonstration of the potential morbidity and mortality caused by an anthrax biological weapon occurred in 1979 following the accidental release of anthrax spores from a production facility in Sverdlosk, Russia. The incident resulted in at least 66 deaths (some sources estimate the death toll to have been greater than 100) due to inhalational anthrax.

Epidemiology and Epizootology

Anthrax is a globally distributed infection. The presence of this disease in humans has been recorded as sporadic, endemic, or hyperendemic in almost every area of the world except the two polar caps, Alaska, and New Zealand. Anthrax is also absent 2,000 m above sea level.

Anthrax is mostly a disease of herbivores, with sheep, goats, cattle, and some other domestic and wild animals typically infected. Anthrax may persist in the environment for many years after contaminating a pasture. Environmental persistence appears to be related to a number of factors, including soil temperature, moisture, and the inherent resistance of anthrax spores to environmental degradation. The world's pastures are permanently infected with anthrax and even if intensive efforts were made to eliminate it in man and domestic animals, it is impossible to eradicate it from the wild.

Human cases of anthrax have traditionally fallen into two categories: agricultural and industrial. Historically, agricultural cases have occurred among laborers in direct contact with infected livestock, while industrial cases tend to involve individuals in contact with infected animal products, particularly workers involved in the processing of wool and meat-bone meal. According to some statistical analyses, the worldwide incidence of anthrax in humans reaches many thousands annually, mostly cutaneous. However, since anthrax is not a reportable disease in many nations, including more than half of the African nations, the true global incidence and prevalence of the disease is unknown. For the past two decades,

the total annual incidence in the United States has been less than one case per year. Most of the U.S. cases in recent decades have resulted from exposure to wool or other animal hair. Before October 2001, the last case of inhalational anthrax in the U.S. occurred in 1978.

Naturally occurring human cases are invariably zoonotic in origin, with no convincing data to suggest that human-to-human transmission has taken place. Of the 235 cases of anthrax reported in the United States between 1955 and 1994, 224 were cutaneous and 11 were inhalational, with 20 fatalities overall.

Epidemics of human anthrax have been reported, the two best described outbreaks being in Zimbabwe between 1978 and 1980 and the accidental release in Sverdlosk, Union of Soviet Socialist Republics (USSR) in 1979. The Zimbabwe epidemic resulted from a cattle outbreak that arose after the breakdown of veterinary care during a civil war. Thousands of human cases occurred in this epidemic. In just one of the provinces, 6500 cutaneous anthrax cases were reported, with approximately 100 fatalities. In Sverdlosk, the great majority of cases were inhalational. Practically all fatalities associated with this outbreak occurred in a 4- to 5-km area downwind from the anthrax production facility.

Three types of anthrax infection occur in humans: inhalational, cutaneous and gastrointestinal. Naturally occurring inhalational anthrax is now a rare cause of human disease. Thus, identification of a case should result in an immediate epidemiologic investigation as to the source of exposure. Cutaneous anthrax is the most common naturally occurring form. The gastrointestinal form, which follows ingestion of undercooked contaminated meat, is uncommon.

The case definition for anthrax, updated in 1996 by the Centers for Disease Control and Prevention (CDC), is as follows:

Clinical description

- An illness with acute onset characterized by several distinct clinical forms, including the following:
- Cutaneous: a skin lesion evolving during a period of 2-6 days from a papule, through a vesicular stage, to a depressed black eschar
- Inhalation: a brief prodrome resembling a viral respiratory illness, followed by development of hypoxia and dyspnea, with radiographic evidence of mediastinal widening
- Gastrointestinal: severe abdominal distress followed by fever and signs of septicemia
- Oropharyngeal: mucosal lesion in the oral cavity or oropharynx, cervical adenopathy and edema, and fever

Laboratory criteria for diagnosis

- Isolation of *Bacillus anthracis* from a clinical specimen, or
- Anthrax electrophoretic immunotransblot (EITB) reaction to the protective antigen and/or lethal factor bands in one or more serum samples obtained after onset of symptoms, or
- Demonstration of *B. anthracis* in a clinical specimen by immunofluorescence:

Case classification

Confirmed: a clinically compatible case that is laboratory confirmed.

Etiology

Bacillus anthracis is a relatively large (1-2 μm), Gram-positive, spore-forming, non-motile, non-flagellated facultative anaerobe. The spores are exceptionally stable under extreme conditions and can survive decades of dormancy. The microorganism grows readily on certain nutrient media, including those containing blood. The processes of sporulation [formation of spores] and capsule formation are observed only when the bacteria are cultivated on certain nutrient media and under certain conditions.

The life cycle of *Bacillus anthracis* has four major phases: vegetative phase (from spores to replicating bacteria), intense growth phase, stationary phase, and sporulation phase. Anthrax spores have a relatively high level of resistance to high temperatures and disinfectants.

Bacillus anthracis forms a poly-D-glutamic acid capsule while in the host animal, when grown with nutrient media containing blood or blood plasma, and in the presence of CO₂. *Bacillus anthracis* does not sporulate in so-called "capsule-forming" media as efficiently as when it is cultivated *in vivo*. According to current knowledge, the capsule is neither immunogenic nor toxigenic, but is considered as a virulence factor as it protects anthrax vegetative cells from the bactericidal components of serum and phagocytosis. The capsule plays its most important role during the establishment of infection and is believed to be less significant in later stages of the disease, which are mediated by the anthrax toxin.

In addition to capsule, virulent strains of *Bacillus anthracis* secrete a set of three distinct antigenic protein components: protective antigen (PA), edema factor (EF), and lethal factor (LF). PA can bind either LF or EF, forming lethal toxin (LeTx) or edema toxin (EdTx). Collectively these two toxins are seen as a complex exotoxin called anthrax toxin. Each component of the toxin is a thermolabile protein with a molecular weight exceeding 80kDa. Edema factor (EF) is an adenylate cyclase that is responsible for the edema seen in anthrax infections. Lethal factor (LF) is a zinc-metalloprotease that is essential for the lethal effect of the anthrax toxin on macrophages. Protective antigen (PA) contains the binding domain of anthrax toxin, which binds to an unidentified receptor on the cell surface and allows translocation of LF or EF into the cell by endocytosis.

Organ and System Pathogenesis

Discovery of anthrax toxins (lethal and edema) in 1955 was a triumph for reductionist thinking, which sought simple causes for the results of complex diseases. It was demonstrated that organisms grown *in vivo* could reveal unknown determinants of bacterial pathogenicity. The toxins were isolated from plasma of guinea pigs dying of anthrax. The mystery seemed finally solved. For the first time major virulence factors of anthrax bacterium were identified. None of the proteins were toxic alone, but lethal factor injected together with protective antigen caused death of experimental animals. The lethal toxin was shown to be essential for virulence of the bacillus because a deletion of the lethal factor gene caused almost a complete loss of virulence. A newly emerging theory for the function of lethal toxin suggests suppression of the proinflammatory response by the infected macrophages early in the infectious process. It seems completely opposite to what was previously believed to be a burst of cytokines and reactive oxygen intermediates. This suppression helps pathogen survival and the intracellular growth of the bacilli critical for the

determination of the infection's outcome. The infected macrophages ultimately die and release bacteria into the lymphatic system where they encounter little defense from the body.

Little is known regarding the late stage conditions of anthrax sepsis and septic shock. Many bacteria that cause sepsis exert pathological effects by triggering an inflammatory response from a number of mediators, including cytokines. The intensity and spectrum of this response in anthrax has not been studied. A contribution of anthrax toxins in this process has long been assumed, but warrants direct investigation. Especially intriguing are the facts of death in macaques immunized against toxin and later challenged with anthrax. In some of the animals a lethal outcome of the infection resulted from very low levels of bacilli in blood, implicating a virulent role of unknown factors distinct from toxins.

The cell wall of the anthrax bacterium may be an important and overlooked factor in anthrax sepsis. While the infection progresses, the availability in host oxygen and nutrients decrease, with a concurrent increase in both bacterial and host metabolic waste products. Death of bacteria at this stage releases a significant amount of free cell wall component into the bloodstream that is expected to result in a massive proinflammatory response. The overproduction of proinflammatory cytokines, caused by stimulation of immune and endothelial cells by anthrax cell wall, coupled with severe hypoxia from the consumption of oxygen by proliferating bacilli, may lead to the multiple organ failure, development of shock and sudden death. This mechanism would explain why antibiotic administration to patients started at late stages of infection is ineffective even though bacteria die in large numbers.

It was shown that anthrax cell wall when used to treat human peripheral blood monocytes induces IL-1 β , TNF- α , and superoxide ion production, activates neutrophils, monocytes and eosinophils, and promotes a mitogenic response from lymphoid cells. In contrast, lethal toxin does not generate comparable activity. Inhalational anthrax initiated in the lungs results in a systemic infection with an extremely high titer of bacilli in the body, including blood, brain and cerebrospinal fluid with little bacterial clearance by the immune system.

Cellular and Molecular Pathogenesis (Inhalational form)

The Toxins

The virulence of *B. anthracis* depends upon three plasmid-encoded factors: two protein toxins and an antiphagocytic poly-D-glutamic acid capsule. The two toxins, collectively called anthrax toxin, are formed by the binary association of three proteins: protective antigen (PA, 83 kilodaltons [kDa]), lethal factor (LF; 90 kDa), and edema factor (EF; 89 kDa). PA can combine with LF to form lethal toxin (LeTx) or edema factor to form edema toxin (EdTx). There is evidence to suggest that calcium plays a significant role in the expression of LeTx activity. Research has shown that removal of calcium from the culture medium inhibits PA binding. It has been suggested that calcium is also required at a step after internalization of lethal toxin.

The cytolytic effect of LeTx is mediated by reactive oxygen intermediates (ROIs). Hanna et al. found that cultured macrophages treated with LeTx release superoxide anion at the onset of lysis. Further, macrophage lysis could be prevented by inhibiting superoxide activity. When LeTx-resistant cell lines were examined, they were found to be deficient in the production of ROIs. These observations suggest that the production of ROIs may also be involved in the lysis of macrophages.

Another factor that plays a significant role in the pathogenesis of anthrax infection is edema toxin (EdTx). EdTx consists of edema factor (EF), which is a calmodulin-activated Ca^{2+} -dependent adenylate cyclase, and PA, the binding moiety that permits entry of the toxin into the host cell. Increased cellular levels of cyclic adenosine monophosphate (AMP) upset water homeostasis and are believed to be responsible for the massive edema. EdTx inhibits neutrophil function in vitro. Interestingly, patients with cutaneous anthrax infection often present with neutropenia.

EdTx has been found to inhibit phagocytosis of opsonized, avirulent *B. anthracis* by polymorphonuclear neutrophils and, more generally, elevated cyclic AMP levels have long been noted to inhibit macrophage phagocytosis. Thus, EdTx might promote bacterial survival during the early stages of infection, thereby acting synergistically with LeTx to promote progression of the infection and death.

The Bacterial Cell Capsule

One more virulence factor that plays an essential role in anthrax infection is the poly-D-glutamic acid capsule, encoded by plasmid pX02. Fully virulent strains of *B. anthracis* have a well-developed polymeric capsule, which inhibits phagocytosis of vegetative anthrax bacilli. It has been determined that capsule formation requires low levels of oxygen and the presence of CO_2 . It has been argued that the *Bacillus anthracis* capsule, like that of many other microorganisms, has neither immunogenic, toxigenic, nor pyrogenic properties. Still, other research argues that the capsule is an important virulence factor, based on the fact that virulent spores germinate within 8 hours and immediately form a capsule, while avirulent spores germinate 50 hours after inoculation. At this time, it is still not clear what role the bacterial capsule plays in anthrax pathogenesis.

Pathologic Anatomy (Inhalational form)

Inhalational anthrax The level of susceptibility of organs and systems in terms of the infection intensity is different. The most susceptible are the lymphatic system, blood and spleen, all of which contain a large proportion of lymphoid tissue.

While *B. anthracis* is in every organ of the body at death, significant concentrations are found in the blood, lungs, and lymph nodes. In fact, the blood is so packed with the bacillus that early researchers believed the cause of death was due to a "log-jam" effect. There is some evidence that *Bacillus anthracis* has a hemolytic effect on the wall of the capillaries, causing widespread hemorrhage. The lungs become massively edematous, atelectic and severe bilateral hydrothorax with up to 2L of fluid in one lung and substantial subpleural and perivascular hemorrhage may occur. Lymph nodes demonstrate profound hemorrhagic necrosis. The mediastinal lymph nodes become black, and nonfunctional. The hilar lymph nodes also become hemorrhagic, with areas of consolidation and necrotic liquefaction. Thoracic hemorrhagic necrotizing lymphadenitis and hemorrhagic necrotizing mediastinitis are common. Bronchioles, pharynx, larynx, and trachea are often covered with thick tenacious mucus. Extraordinarily large amounts of pleural effusion are present.

Localized hemorrhage occurs in the gastrointestinal tract. There are circumscribed mucosal erosions and submucosal hemorrhage. Neutrophils have been identified in the serosal layer of the stomach. Subarachnoid hemorrhage has been found over the entire cerebrum along with hemorrhage in the Virchow-Robin space, in the lateral ventricle, the cerebellum, and in the brainstem. There is hemorrhage on the tips of villi in the small intestine.

Clinical Manifestations

Disease types and their details are given below:

1) Inhalational Anthrax

- Incubation Period : 1-6 days
- Prodromal Period : 2-3 days
- Manifestation Period : 1-3 days
- Period of Outcome : Hours

2) Gastrointestinal Anthrax

- Incubation Period : 1-5 days
- Prodromal Period : None
- Manifestation Period : N/A
- Period of Outcome : N/A

3) Cutaneous Anthrax

- Incubation Period : 1-5 days
- Prodromal Period : None
- Manifestation Period : 1-2 days
- Period of Outcome : 7-14 days

4) Oropharyngeal Anthrax

- Incubation Period : 2-5 days
- Prodromal Period : None
- Manifestation Period : N/A
- Period of Outcome : N/A

Patients may exhibit a combination of some of these symptoms, some more commonly than others. The symptoms are ranked from highly likely to rare:

INHALATIONAL ANTHRAX

General Symptoms - fever, pain, diaphoresis, headache, general myalgias, backache, chills, fatigue, malaise, weakness, lethargy

Pulmonary - cough, sore throat, chest pain, shortness of breath, dyspnea, orthopnea, presence of sputum or nasal discharge, decreased breath sounds, increased rate of respirations, respiratory distress syndrome. Chest and breath sounds include egophony, wheezing, and stridor. On examination, rales, dullness to percussion, rhonchi may be noted.

Neurologic - agitation, confusion, disorientation, neck rigidity, Kernig's sign, Brudzinski's sign, syncope, coma. Muscle reflexes as Babinski reflex, plantar reflex, deep tendon reflexes, flaccid paralysis, spasticity of extremities. Eye involvement as photophobia, redness of conjunctivae, distortion of visual field, excessive lacrimation without inflammation, nystagmus, conjugate deviation of eyes.

Circulatory - significant tachycardia, cyanosis, hypotension, hypoxia, dyscrasia

Gastrointestinal - vomiting, nausea, generalized abdominal pain, anorexia

Dermatologic - non-pitting edema, mottled appearance, clammy skin

GASTROINTESTINAL ANTHRAX

A combination of above symptoms (except pulmonary), the most prominent being severe abdominal pain, and may have bloody diarrhea.

CUTANEOUS ANTHRAX

A mild combination of above symptoms along with the presence of black eschars.

Onset : abrupt

Duration : days

Incubation : 1-6 days

Differential Diagnosis

Inhalational

The initial symptoms of inhalational anthrax are nonspecific or "flu-like" and are similar to those of atypical pneumonia from other causes. The prognosis is improved if early treatment is implemented, so that a high level of suspicion is necessary. This suspicion is based on epidemiological clues from the patient's history as well as clinical signs and symptoms.

Distinguishing inhalational anthrax from influenza or influenza-like illness (ILI) may be difficult in the early stages of illness, but experience with the inhalational anthrax cases that occurred in October 2001 suggests differences in signs and symptoms that may be helpful in evaluating the patient clinically. Fever, fatigue, and cough are common for all three, but almost all of the patients with inhalational anthrax also reported nausea and vomiting and shortness of breath. These are uncommon symptoms with influenza or ILI. Also, rhinorrhea was uncommon among the anthrax cases but is a typical symptom with "flu-like" illnesses. A key finding was that all of the anthrax cases demonstrated radiographic changes, which is not typical in uncomplicated influenza or ILI. Rapid testing for influenza is available and may assist with diagnosis. The clinician should also take into account the types of illness that have been occurring in the community.

Anthrax infection is unusual in that mediastinal changes can be detected early in the course of infection by chest radiography. However, a similar picture can be seen in acute bacterial mediastinitis and fibrous mediastinitis due to *Histoplasma capsulatum*. Less specific findings include pleural effusions and radiographic evidence of pulmonary edema. Silicosis, siderosis, alveolar proteinosis, and sarcoidosis are often alternative causes of chronic mediastinitis in patients with the relevant occupational history and previous chest radiographs demonstrating long-standing mediastinal widening.

Cardiopulmonary collapse associated with a radiographic history of mediastinal widening in the late stages of inhalational anthrax must be differentiated from cardiovascular collapse of noninfectious origins. Noninfectious causes of cardiovascular collapse due to mediastinal widening include dissecting or ruptured aortic aneurysm and esophageal rupture.

Hemorrhagic meningitis caused by anthrax must be distinguished from subarachnoid hemorrhage by computer tomography without contrast. To distinguish hemorrhagic meningitis caused by *B. anthracis* from that caused by other bacteria, Gram staining and culture of cerebrospinal fluid should be performed. In addition to the above indicators, the clinician should consider anthrax if there is a history of contact with materials that may be contaminated with spores, such as infected farm animals and imported hides, or travel to places where anthrax is endemic.

Gastrointestinal

When ingestion of contaminated meat is suspected, the symptoms of an acute abdomen should be considered as possible early signs of gastrointestinal anthrax infection.

Cutaneous

In cutaneous anthrax, the painless, blackened, necrotic eschar is limited to the late stage of the infection. The ulcerative eschar of cutaneous anthrax must be differentiated from other papular lesions that present with regional lymphadenopathy. If the lesion is purulent and the regional lymph nodes are palpable, staphylococcal lymphadenitis is the most likely cause, although cutaneous anthrax lesions can be super-infected with pyrogenic bacteria. The differential diagnosis should include tularemia, staphylococcal or streptococcal disease.

Diagnosis

The most critical aspect in making a diagnosis of anthrax is a high index of suspicion associated with a compatible history of exposure.

Inhalational Anthrax

The diagnosis of inhalational anthrax is extraordinarily difficult, but the disease should be suspected with a history of exposure to a *B. anthracis*-containing aerosol. The first evidence of a clandestine release of anthrax as a biological weapon most likely will be patients seeking medical treatment for symptoms of inhalational anthrax. The sudden appearance of a large number of patients in a city or region with an acute-onset flu-like illness and case fatality rates of 80% or more, with nearly half of all deaths occurring within 24 to 48 hours, is highly likely to be anthrax.

The early symptoms are entirely nonspecific. However, (1) the development of respiratory distress in association with radiographic evidence of a widened mediastinum due to hemorrhagic mediastinitis, and (2) the presence of hemorrhagic pleural effusion or hemorrhagic meningitis should suggest the diagnosis. Sputum examination is not helpful in making the diagnosis, since pneumonia is not usually a feature of inhalational anthrax.

Gastrointestinal Anthrax

Gastrointestinal anthrax is exceedingly difficult to diagnose because of the rarity of the disease and its nonspecific symptoms. Only with a history of ingesting contaminated meat in the setting of an outbreak is diagnosis usually considered. Microbiologic cultures are not helpful in confirming the diagnosis. The diagnosis of oropharyngeal anthrax can be made from the clinical and physical findings in a patient with the appropriate epidemiological history.

Cutaneous Anthrax

Cutaneous anthrax should be considered following the development of a painless pruritic papule, vesicle, or ulcer-often with surrounding edema-that develops into a black eschar. With extensive or massive edema, such a lesion is almost pathognomonic. Gram stain or culture of the lesion will usually confirm the diagnosis.

Meningitis

Meningitis due to anthrax is clinically indistinguishable from meningitis due to other etiologies. An important distinguishing feature is that the cerebral spinal fluid is hemorrhagic in as many as 50% of cases. The diagnosis can be confirmed by identifying the organism in cerebrospinal fluid by microscopy, culture, or both.

Laboratory Diagnosis

Specimens for culture should be obtained from a malignant pustule, sputum, or blood. A Gram stain and a fluorescent-antibody stain are useful in making a presumptive diagnosis. The organism will grow readily on most laboratory media. However, the distinction between *B. anthracis* and nonpathogenic bacilli such as *B. cereus* present the greatest difficulty for laboratory diagnosis. This is complicated by the fact that most laboratory personnel have never seen *B. anthracis*.

The Ouchterlony gel diffusion, microhemagglutination, and enzyme-linked immunosorbent assay (ELISA) procedure can demonstrate antibodies to the organism. Acute and convalescent sera of suspect cases should be obtained and submitted to the Centers for Disease Control and Prevention for confirmation of infection.

New diagnostic techniques have focused on the use of the polymerase chain reaction to amplify markers specific to *B. anthracis* or the *B. cereus* group. Two markers, *vrrA* and *Ba813*, have been the subject of extensive study. Other methods using the polymerase chain reaction to amplify specific virulence plasmid markers harbored by different anthrax strains may soon become available.

Morbidity and Mortality

All forms of anthrax can be lethal if not treated. The types of anthrax and mortality information is given below:

1) Cutaneous Anthrax

- Mortality with antibiotics is about 1%
- Septicemia is very rare

2) Gastrointestinal Anthrax

- Mortality : 50-100%
- Morbidity is due to blood loss, fluid and electrolyte imbalance and subsequent shock.
- Death results from intestinal perforation or anthrax toxemia.

3) Inhalation Anthrax

- Mortality is 95-100%
- Death is universal in untreated cases and may occur in as many as 95% of treated cases if the therapy is begun more than 24-48 hours after the onset of symptoms

Vaccination

An FDA licensed vaccine, derived from the supernatant fluid of an attenuated, nonencapsulated *B. anthracis* strain (Stern) is available and has been used in hundreds of thousands of military troops and at-risk civilians. The vaccine, designated "anthrax vaccine adsorbed" (AVA) is non-living and the primary antigen is a protein called protective antigen. The vaccination series, as currently licensed, consists of six doses (0,2 and 4 weeks and 6, 12 and 18 months) followed by annual boosters. AVA is administered subcutaneously as a 0.5-ml dose. Efforts are underway to reduce the number of doses now required by the package insert. There are not enough data from exposure of humans to determine protective efficacy of the vaccine against aerosol challenge, but studies in rhesus monkeys indicate that the vaccine is effective, even when as few as two doses are administered. The U.S. Army has also developed a next generation anthrax vaccine, based on the same antigen (PA) produced through recombinant technologies. Although there is no reason to believe that the new vaccine will be more protective, it will be more easily produced in available production facilities and may be slightly less reactogenic and possibly less costly if large lots are needed.

Individuals exposed, or potentially exposed, to the "anthrax letters" during October 2001 were put on antibiotics for 30-60 days. Subsequently, they were offered the vaccine. Recently, after considering the ease of attack, small size of the payload and vulnerability of millions of Americans in highly populated areas, some experts have begun to reconsider the earlier conclusion that immunizing the entire population might not be feasible. For now, it would not be unreasonable for essential service personnel to avail themselves of the vaccine, should it become available.

The safety of AVA has been the subject of numerous studies. The Institute of Medicine recently published a report which concluded that AVA is effective against inhalational anthrax and may help prevent onset of disease post-exposure, if given with appropriate antibiotics. The IOM committee also concluded that the vaccine was acceptably safe. At the U.S. Army Medical Research Institute of Infectious Diseases, where 1,583 humans had received 10,722 doses over a 25 year period, 1% of inoculations were associated with one or more systemic events (headache---most common at 0.4% of doses---malaise, myalgia, fever, nausea, vomiting, dizziness, chills, diarrhea, hives, anorexia, arthralgias, diaphoresis,

blurred vision, generalized itching or sore throat) and 3.6% with local reactions. No long-term sequelae were reported.

Post-Exposure Prophylaxis

Post-exposure prophylaxis against anthrax may be achieved with oral ciprofloxacin (500 mg orally q 12 h) or doxycycline (100 mg orally q 12 h), and all persons exposed to a bioterrorist incident involving anthrax should be administered one of these regimens at the earliest possible opportunity. Adherence to the antibiotic prophylaxis program must be strict as disease can result at any point within 30-60 days after exposure, if antibiotics are stopped. In case of threatened or suspected release of anthrax, chemoprophylaxis can be delayed 24 to 48 hours, until the threat is verified. Chemoprophylaxis can be discontinued if the threat is found false. Levofloxacin and ofloxacin would be acceptable alternatives to ciprofloxacin. In November 2002, the Advisory Committee on Immunization Practices released supplemental recommendations for the use of anthrax vaccine in addition to post-exposure chemoprophylaxis for patients potentially exposed to inhalational anthrax. A three dose regimen at 0, 2, and 4 weeks post exposure is recommended in addition to chemoprophylaxis as detailed above.

Anti-microbial treatments

Penicillin has historically been the drug of choice for treatment of naturally occurring strains of anthrax, with ciprofloxacin and doxycycline suitable alternatives. However, the possibility of penicillin resistance should be suspected in a biological warfare attack. Cutaneous anthrax may be treated orally, while gastrointestinal or inhalational disease should receive high doses of intravenous antibiotics (ciprofloxacin 400 mg q 12 h or doxycycline 100 mg q 12 h plus one or two additional antimicrobials as listed below*). Begin IV treatment initially. Change to oral antibiotic therapy when clinically appropriate (ciprofloxacin 500 mg po BID or doxycycline 100 mg po BID.) Therapy should continue for 60 days IV and po combined. The more severe forms will require intensive supportive care.

The pediatric dosages are ciprofloxacin 10-15mg/kg IV q 12h dosing up to 1 Gm/day OR doxycycline based on age and weight as follows: >8 yrs and >45 kg, 100 mg IV q 12h; >8 yrs and ≤45 kg, 2.2mg/kg IV q 12h; ≤8yrs, 2.2mg/kg IV q 12h. The oral dosages are ciprofloxacin 10-15 mg/kg po q 12, OR Doxycycline as follows: >8 yrs and >45 kg, 100 mg po BID; >8 yrs and ≤45 kg, 2.2mg/kg po BID; ≤8yrs, 2.2mg/kg po BID.

Additional information regarding antimicrobial therapy is available from the CDC as published in the [MMWR, 10/26/2001; 50\(42\), 909-919](#)

* Rifampin, Vancomycin, Penicillin, Ampicillin, Chloramphenicol, Imipenem, Clindamycin, Clarithromycin, Azithromycin, Cefotaxime, Ceftriaxone

Protection/Isolation/Notification Measures

Protection

According to the Centers for Disease Control and Prevention (CDC) Biosafety in Microbiological and Biomedical Laboratories manual, 4th edition, Biosafety Level 2 practices, containment equipment, and facilities are recommended for activities using clinical materials and diagnostic quantities of infectious cultures. Animal Biosafety Level 2 practices,

containment equipment, and facilities are recommended for studies utilizing experimentally infected laboratory rodents. Biosafety Level 3 practices, containment equipment, and facilities are recommended for work involving production quantities or concentrations of cultures and for activities - such as centrifugation -- with a high potential for aerosol production.

Isolation

Anthrax infection is not contagious and cannot be transmitted by human-to-human contact. Therefore no isolation is necessary. (Note: Standard precautions are still indicated for patient care as part of routine infection control practice.) Cadavers should not be opened within 30 minutes of death as the bacteria sporulate immediately on contact with ambient air.

Notification

Anthrax is a notifiable disease in all 50 states and is federally reportable. According to bioterrorism guidelines put forth by CDC, any case of inhalational (pulmonary) anthrax in the U.S. should also be reported to the Federal Bureau of Investigation (FBI), as it is assumed that inhalational anthrax is so rare in the United States that any case must be due an intentional release.

Please visit the following link to the CDC Website for city and county health department contact information, as well as emergency hotline numbers.

Sources For Further Information

The CDC [Bioterrorism Preparedness and Response](#) Website

The Johns Hopkins University Center for Civilian Biodefense Studies
(<http://www.hopkins-biodefense.org>)

Emerging Infectious Disease Journal [Vol. 5 No. 4 1999.](#)

CDC Division of Bacterial and Mycotic Disease [Fact Sheet on Anthrax](#)

Emerging Infectious Diseases, Vol. 8 No. 10, October 2002

<http://jama.ama-assn.org/issues/v281n18/ffull/jst80027.html>

<http://www.ama-assn.org/ama/pub/category/6631.html>

<http://www.hopkins-biodefense.org/pages/agents/agentanthrax.html>

<http://www.who.int/emc/diseases/anthrax/faqanthrax.html>

http://www.who.int/emc-documents/zoonoses/whoemczdi986c.html#_Toc433123882

<http://www.ericse.org/anthrax.html> (very detailed links)

http://www.vetmed.lsu.edu/whocc/mp_world.htm

<http://www.nlm.nih.gov/medlineplus/anthrax.html>

<http://www.fda.gov/cber/vaccine/anthrax.htm>

<http://www.niaid.nih.gov/newsroom/releases/anthrax.htm>

<http://www.aphis.usda.gov/oa/pubs/anthrax.html>

<http://usinfo.state.gov/topical/global/hiv/anthrax.htm>

<http://www.hhs.gov/hottopics/healing/biological.html>
<http://www.hhs.gov/news/press/2001pres/20011010a.html>
<http://www.epa.gov/epahome/hi-anthrax.htm>
<http://www.whitehouse.gov/homeland/anthrax-faq.html>
http://science.nasa.gov/headlines/y2002/01feb_anthrax.htm (air cleaning device)
<http://www.fbi.gov/pressrel/pressrel01/mueller101601.htm> (photos of letters)
http://wire.ap.org/APpackages/anthrax_flash/
<http://www.nbc-med.org/SiteContent/HomePage/WhatsNew/MedAspects/Ch-22electrv699.pdf>
<http://www.aap.org/advocacy/releases/anthraxqa.htm> (american academy of pediatrics)
<http://www.merck.com/pubs/mmanual/section13/chapter157/157c.htm>
<http://www.redcross.org/news/ds/0109wtc/anthrax/anthraxbro.pdf>
<http://www.gwu.edu/~nsarchiv/NSAEBB/NSAEBB61/>(chapter from book links to real docs)
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