Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

CHAPTER 80

Future Biological and Chemical Weapons

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HISTORICAL PERSPECTIVE

Biological and chemical weapons have been used throughout history.[1] For millennia, indigenous South American peoples deliberately used plant-derived arrow poisons such as curare and toxins from poison dart frogs, although these preparations were used mainly for hunting. Similar toxins were used in Africa. The ancient Greeks, for whom *toxikon* meant "arrow poison," tipped arrows with winter aconite, and this practice continued into medieval Europe, persisting into the seventeenth century in Spain and Portugal.[2] Soldiers in India used smoke screens, incendiary weapons, and toxic fumes as early as 2000 BCE, and the Sung Dynasty in China employed a wide variety of arsenical smokes and other poisons in battle. The military use of toxins dates from at least the sixth century BCE, when Assyrian soldiers poisoned enemy wells with ergot-contaminated rye. In 423 BCE, during the Peloponnesian War, Thracian allies of Sparta captured the Athenian fort at Delium by using a long tube and bellows to blow poisonous smoke from coals, sulfur, and pitch into the fort. Greek fire (likely composed of rosin, sulfur, pitch, naphtha, lime, and saltpeter) was invented in the seventh century CE and proved to be a very effective naval weapon. Various poisons saw battlefield use during medieval times, and the use of poisons for murder (including assassinations) became widespread. Other examples before the twentieth century include the contamination of water by dumping the corpses of dead humans or animals into wells, the use of snakes and other creatures as poisonous vectors, and occasionally, fomites to transmit infections such as smallpox to unsuspecting victims. This latter technique was used with remarkable success during the French and Indian War (1754-1767), when Sir Jeffrey Amherst was alleged to have given "gifts" (blankets) harboring the pus and scabs from smallpox victims to unsuspecting Native Americans. The Indians possessed no immunity against smallpox and thus experienced very high rates of infection and mortality as smallpox swept through the local tribes.[3]

During the late nineteenth and early twentieth centuries, the science and technology necessary for the development of sophisticated biological and chemical weapons proceeded apace. World War I saw the first large-scale use of "poison gas," including lacrimators, chlorine, phosgene, arsenicals, cyanide, and sulfur mustard. By the end of the war, nearly one in every three rounds was a chemical munition. Dr. Shiro Ishii and other Japanese scientists in the infamous Unit 731 worked on the weaponization of anthrax, plague, smallpox, and tetrodotoxin as well as a variety of chemical agents during World War II. There are even suspicions that the bomb used in the assassination of Reinhard Heydrich in Czechoslovakia in 1942 contained botulinum toxin.[4] After World War II, ricin was used as an injectable assassination weapon, and in the 1970s and 1980s T-2 toxin, a trichothecene mycotoxin, was alleged to have been the toxic component of the "yellow rain" employed against H'Mong refugees from Laos. More recently, Iraq and Iran both used chemical weapons against each other in the Iran-Iraq War of the 1980s, and Iraq had a weapons program that included the development of sulfur mustard, nerve agents, "Agent 15" (an anticholinergic incapacitating agent), botulinum toxin, epsilon toxin from *Clostridium perfringens*, and aflatoxin.[5] Militia groups in the United States and terrorist groups throughout the world have used ricin for political purposes.

American scientists started developing chemical weapons as a response to the use of chemical warfare in Europe during World War I and conducted both offensive and defensive research on biological and chemical weapons. However, in 1969 the United States unilaterally renounced the first use of chemical agents, halted chemical-agent production, and terminated its offensive biological weapons program.

In 1972 the Biological Weapons and Toxins Convention was created; it was signed by representatives from 104 nations, including the United States (which ratified the Convention in 1975), the Soviet Union, and Iraq, although many signatories did not consider toxins to be biological weapons and did not consider the treaty binding on toxin use. Since that time, at least 140 nations have either signed or ratified this treaty.[6] However, the Soviet Union and Iraq began violating the treaty in short order. In the Soviet Union, weapons scientists stepped up research and development of numerous biological and chemical weapons as part of one of the largest and most comprehensive biological-weapons programs in history. Soviet scientists created large stockpiles of weaponized anthrax, plague, smallpox, tularemia, nerve agent, mustard, and other biological and chemical agents.[5]

In 1979 the world was put on notice of the devastating potential that biological weapons pose to humanity. In that year, a small quantity of weapons-grade anthrax was accidentally released from a manufacturing plant located in the former city of Sverdlovsk (now Yekaterinburg) in Russia, resulting in 77 cases and 66 deaths. Dr. Matthew Meselson, a Harvard scientist, was permitted to study the event many years later and reported the results of his work in a 1979 *Science* article. Meselson determined that the majority of the deaths had occurred among victims living in a narrow, 4-km-wide band downwind from the plant. Animal deaths were confirmed as far as 30 km downwind. Meselson further concluded that less than 1 g of weapons-grade anthrax had been released from the plant.[7] If his calculations are accurate, weaponized anthrax possesses staggering potential as a biological weapon given its stability, its relative ease of production, and its ability to be dispersed in a clandestine manner over great distances.

In March 1995, after having unsuccessfully attempting to deploy biological agents, members of the Aum Shinri Kyo cult executed a coordinated attack with the nerve agent sarin (GB) on the Tokyo subway system. More than 5500 people sought medical treatment, and a dozen died. The Aum Shinri Kyo had used sarin in Matsumoto 9 months earlier in an attack that had exposed more than 300 people and killed 7 in an attempt to assassinate judges unfavorable to their cause.[8,9]

The anthrax attacks in the fall of 2001 involved the use of letters containing weapons-grade anthrax mailed through the U.S. postal system. Five people died, and 17 became ill with either cutaneous or inhalational anthrax. Buildings contaminated with spores included the Hart Senate Office building and the Brentwood postal facilities in Washington, DC. It cost millions of dollars to rehabilitate these buildings. The anthrax used in the attacks was determined to be extremely potent and could have caused far greater numbers of casualties had it been dispersed more widely.[10,11]

The use of chemical weapons also occurred in recent history. In September 2013 the United Nations (UN) released their investigations on the use of chemical weapons in Syria. The UN concluded that sarin gas was used on August 21, 2013, in the Ghouta area of Damascus against "civilians, including children, on a relatively large scale."[12] These findings were based on interviews with survivors and other witnesses, documentation of munitions and their components, collection of environmental samples for subsequent analysis, assessment of symptoms of survivors, and collection of hair, urine, and blood samples.

According to Dr. Ken Alibek, former Deputy Director of Biopreparat, the Soviet Union's nominally civilian medical research institute, Soviet scientists and physicians spent large sums of money and manpower during the 1980s and 1990s developing the most lethal and potent biological weapons known to man. In addition to weaponizing the etiologic agents of anthrax, smallpox, Marburg fever, and others, they created antibiotic-resistant strains of *Yersinia pestis* (plague), *Francisella tularensis*, and other pathogens. Furthermore, by applying genetic engineering techniques, the Soviets are also alleged to have created pathogens with novel characteristics and strains of several organisms capable of defeating certain vaccines.[13]

As we enter the biotechnological revolution of the twenty-first century, our understanding of molecular biology, genetics, and biochemistry is exploding. The human genome has been sequenced, and it is now possible to manipulate genes from disparate organisms to create new and novel pathogens. Scientists are also able to synthesize and weaponize a number of different endogenous biological-response modifiers including cytokines, hormones, neurotransmitters, and plasma proteases. But even nature continues to surprise us. New, naturally occurring infections with the potential to cause large-scale human diseases and death continue to emerge at an ever-increasing rate throughout the world, and it is conceivable that these pathogens could also be weaponized by enterprising scientists.

This chapter briefly reviews the future of chemical and biological weapons as we enter this new era of explosive growth in our understanding of the life sciences. We are presented with an extraordinary opportunity to solve a host of human afflictions or to create new classes of biological and chemical weapons that have the capacity to destroy our civilization as we know it today.

FUTURE BIOLOGICAL WEAPONS

The appearance of a new or reemerging infectious disease has global implications. During the past 20 years, more than 30 new lethal pathogens have been identified.[14] A classic example of this emerging threat is pandemic influenza. In 1918, as World War I was coming to an end, the Spanish flu struck with devastating consequences. In less than 1 year, this virus was able to circumnavigate the globe and kill an estimated 40 million people.[15] More recently, the emergence of severe acute respiratory syndrome (SARS) in Southeast Asia resulted from a coronavirus that jumped species from animals to humans and rapidly spread to 29 countries in less than 90 days. Finally, the 2014 outbreak of Ebola in Western Africa, still raging as of this writing, is an example of how devastating these agents can be when they emerge in a region previously naïve to them. Novel and dormant infectious agents such as SARS, influenza, and Ebola appear to be emerging or reemerging with increasing frequency and with greater potential for serious consequences. Many factors contribute to the emergence of new diseases: environmental changes, global travel and trade, social upheaval, and genetic changes in infectious agent, host, or vector populations. Once a new disease is introduced into a suitable human population, it often spreads rapidly and has a devastating impact on the medical and public health infrastructure. If the disease is severe, it may lead to social disruption and have a profound economic impact. Outbreaks of emerging or reemerging diseases may be difficult to distinguish from outbreaks resulting from intentional introduction of infectious diseases for nefarious purposes.

As scientists develop more sophisticated laboratory procedures and increase their understanding of molecular biology and the genetic code, the possibility of bioengineering more virulent, antibiotic, and vaccine-resistant pathogens for military or terrorist uses becomes increasingly likely. It is already theoretically possible to synthesize and weaponize certain biological response modifiers (BRMs) as well as to engineer genomic weapons capable of inserting novel DNA into host cells. The potential to cause widespread disease and death with any of these weapons is incalculable and concerning. Scientists and policy makers have begun to address the issue with a robust research agenda to develop medical countermeasures.

Ebola hemorrhagic fever, as of December 4, 2014, had caused 6055 deaths among 17,111 confirmed cases in western Africa. Scientists have debated whether Ebola could be weaponized into a weapon of mass destruction by terrorists. The consensus seems to be that this would be a very difficult undertaking because of the knowledge and laboratory skills that are required. Moreover, the biology of the virus does not lend itself well to weaponization.[16] Of course, nefarious individuals could use Ebola in a number of ways, in much the same way that a suicide bomber straps on a vest laden with explosives. In theory a person could deliberately infect themselves with the virus and then attempt to infect others once they become symptomatic.

Existing Agents and Their Potential for Future Use

Important existing biological agents with the potential for weaponization for military or terrorist use include the following:

1. Biological agents
   a. *Bacillus anthracis* (anthrax; see Chapter 124)
   b. *Yersinia pestis* (plague; see Chapter 125)
   c. *Francisella tularensis* (tularemia[17]; see Chapter 126)
   d. *Brucella* species (brucellosis; see Chapter 127)
   e. *Coxiella burneti* (Q fever; see Chapter 128)
   f. *Rickettsia prowazekii* (typhus fever; see Chapter 129)
   g. *Orientia tsutsugamushi* (scrub typhus; see Chapter 130)
   h. *Rickettsia rickettsii* (Rocky Mountain Spotted Fever; see Chapter 131)
   i. *Vibrio cholerae* (cholera; see Chapter 132)
   j. *Shigella dysenteriae* (shigellosis; see Chapter 133)
   k. *Salmonella* species (salmonellosis; see Chapter 134)
   l. *Salmonella typhi* (typhoid fever; see Chapter 135)
   m. *Burkholderia mallei* (glanders; see Chapter 136)
   n. *Burkholderia pseudomallei* (melioidosis; see Chapter 137)
   o. *Chlamydia psittaci* (psittacosis; see Chapter 138)
   p. *Escherichia coli* O157:H7 (hemorrhagic *E. coli*; see Chapter 139)
2. Viral agents
   a. Viral encephalitides (alphaviruses; see Chapter140)

   b. Tick-borne encephalitis virus (see Chapter 141)
   c. Viral hemorrhagic fever viruses (arenaviruses, bunyaviruses, filoviruses, flaviviruses; see Chapters 142-145)
   d. Chikungunya virus (see Chapter 146)
   e. Variola major virus (smallpox; see Chapter 147)
   f. Influenza virus (see Chapter 148)
   g. Monkeypox (see Chapter 149)
   h. Hantavirus pulmonary syndrome (see Chapter 150)
   i. Henipavirus (Hendra virus and Nipah virus encephalitis; see Chapter 151)
   j. SARS-CoV (see Chapter 152)
3. Toxins
   a. Staphylococcal enterotoxin B (see Chapter 153)
   b. *Clostridium botulinum* toxin (botulism; see Chapter 154)
   c. *Clostridium perfringens* toxin (epsilon toxin; see Chapter 155)
   d. Marine toxin (see Chapter 156)
   e. T-2 toxin (trichothecene mycotoxins; see Chapter 157)
   f. Ricin toxin from *Ricinus communis* (castor beans; see Chapter 158)
   g. Aflatoxin (*Aspergillus* species; see Chapter 159)
4. Other biological agents
   a. *Coccidioides immitis* (coccidioidomycosis; see Chapter 160)
   b. *Histoplasma capsulatum* (histoplasmosis; see Chapter 161)
   c. *Cryptosporidium parvum* (cryptosporidiosis; see Chapter 162)

Another way to view the relative importance of the above list of agents and diseases list is to consider The Centers for Disease Control and Prevention (CDC) strategy. The CDC categorizes bioterrorism agents or diseases as category A, B, or C.[17] *Category A* agents pose the highest risk to the public and are characterized as follows:

- Easily disseminated or transmitted from person to person
- Can cause high mortality rates and possess the potential for profound public impact
- Could cause public panic and social disruption
- Require special preparations for adequate public health preparedness

Diseases and agents

- Anthrax (*Bacillus anthracis*)
- Botulism (*Clostridium botulinum* toxin)
- Plague (*Yersinia pestis*)
- Smallpox (variola major)
- Tularemia (*Francisella tularensis*)
- Viral hemorrhagic fever: filoviruses (e.g., Ebola and Marburg) and arenaviruses (e.g., Lassa and Machupo)

Category B agents are the next highest priority and are characterized by

- Moderately easy to disseminate
- Cause moderate morbidity and low mortality
- Require specific enhancements of CDC's diagnostic capacity and enhanced disease surveillance

Diseases and agents

- Epsilon toxin of *Clostridium perfringens*
- Food safety threats (*Salmonella* species, *Escherichia coli* O157:H7, *Shigella*)
- Glanders (*Burkholderia mallei*)
- Melioidosis (*Burkholderia pseudomallei*)
- Psittacosis (*Chlamydia psittaci*)
- Q fever (*Coxiella burnetii*)
- Ricin toxin (*Ricinus communis*—castor beans)
- Staphylococcal enterotoxin B
- Typhus fever (*Rickettsia prowazekii*)
- Viral encephalitis (alphaviruses: Venezuelan equine encephalitis, eastern equine encephalitis, western equine encephalitis)
- Water safety threats (*Vibrio cholerae*, *Cryptosporidium parvum*)

Category C agents form the third highest priority and include emerging pathogens that could be engineered for mass dissemination in the future because of the following:

- Availability
- Ease of production and dissemination
- Potential for high morbidity and mortality rates and major health impact

Agents

- Emerging and reemerging infectious diseases such as Nipah virus, hantavirus, human influenza, avian influenza, SARS and SARS-associated coronavirus (SARS-CoV), and Middle East respiratory syndrome (MERS)

Selected Emerging and Reemerging Infections with Weaponization Potential

Because emerging diseases are so diverse and endemic to different geographic locations, their complete description is beyond the scope of this chapter. However, some of these infections may become future threats as agents of biological warfare or terrorism. The most worrisome emerging infectious disease may well be the one we do not know about. Recent experience with HIV, Ebola hemorrhagic fever, SARS, monkey pox, West Nile fever, and hundreds of other "new" diseases reveal that we will continue to be surprised.

Avian Influenza

Avian influenza, or highly pathogenic avian influenza, has periodically caused human infections primarily through close contact with avian species, most often through occupational contact at chicken or duck farms in Southeast Asia. A large outbreak of avian influenza involving the H5N1 strain and human cases occurred in 2004 and originated in two countries from this region.[18] No sustained human-to-human transmission was reported, but there is some evidence that isolated episodes did occur, and the potential exists for genetic reassortment between avian and human or animal strains of influenza. A recent report in the journal *Science* linked the influenza virus responsible for the 1918 epidemic to a possible avian origin.[19] If true, avian influenza may pose a much greater danger to human populations than previously reported. The disease presents in humans in a fashion similar to other types of influenza viruses. It usually begins with fever, chills, headaches, and myalgias and often involves the upper and lower respiratory tract with development of cough, dyspnea, and, in severe cases, acute respiratory distress syndrome. Laboratory findings may include pancytopenia, lymphopenia, elevated liver enzymes, hypoxia, a positive reverse transcriptase-polymerase chain reaction (RT-PCR) test for H5N1, and a positive neutralization assay for H5N1 influenza strain. in vitro studies suggest that the neuraminidase (NA)-inhibitor class of drugs may have clinical efficacy in the treatment and prevention of avian influenza infection.[20]

Human Influenza

The threat for pandemic spread of human influenza viruses is substantial. The pathogenicity of human influenza viruses is directly related to their ability to alter their eight viral RNA segments rapidly; the new antigenic variation results in the formation of new hemagglutinin (HA) and NA surface glycoproteins, which may go unrecognized by an immune system primed against heterologous strains.

Two distinct phenomena contribute to a renewed susceptibility to influenza infection among persons who have had influenza illness in the past. Clinically significant variants of influenza A viruses may result from mutations occurring in the HA and NA genes and expressed as

minor structural changes in viral surface proteins. As few as four amino acid substitutions in any two antigenic sites can cause such a clinically significant variation. These minor changes result in an altered virus able to circumvent host immunity. Moreover, genetic reassortment between avian and human or avian and porcine influenza viruses may lead to the major changes in HA or NA surface proteins known as antigenic shift. In contrast to the gradual evolution of strains subject to antigenic drift, antigenic shift occurs when an influenza virus with a completely novel HA or NA formation moves into humans from other host species. Global pandemics result from such antigenic shifts.

Influenza causes in excess of 30,000 deaths and more than 100,000 hospitalizations annually in the United States. Pandemic influenza viruses have emerged regularly in 10- to 50-year cycles for the last several centuries. During the last century, influenza pandemics occurred 3 times: in 1918 ("Spanish influenza," an H1N1 virus), in 1957 (Asian influenza, an H2N2 subtype strain), and in 1968 (Hong Kong influenza, an H3N2 variant). The 1957-1958 pandemic caused 66,000 excess deaths, and the 1968 pandemic caused 34,000 excess deaths in the United States. The 1918 influenza pandemic illustrates a worst-case public health scenario; it caused 675,000 deaths in the United States and 20 to 40 million deaths worldwide.[19] Morbidity in most communities was between 25% and 40%, and the case-mortality rate averaged 2.5%. A reemergent 1918-like influenza virus would have tremendous societal effects, even in the event that antiviral medications were effective against this more lethal influenza virus.

SARS and SARS-Associated Coronavirus

SARS-associated coronavirus (SARS-CoV) emerged as the cause of SARS during 2003. That year, SARS was responsible for approximately 900 deaths and more than 8000 infections in people from at least 29 countries worldwide. Before a case definition had been clearly established, Chinese authorities reported to the World Health Organization (WHO) more than 300 cases of an atypical pneumonia with 5 related deaths, all originated from Guangdong province in China during February 2003. The infection quickly spread as infected patients traveled to Hong Kong and from there to Vietnam, Canada, and other locations. Only eight laboratory-confirmed cases occurred in the United States, but there is concern that the U.S. population is vulnerable to a widespread outbreak of SARS such as the one that occurred in China, Hong Kong, Singapore, Toronto, and Taiwan in 2003.[21]

A SARS case definition evolved from this initial report to the WHO by Chinese health authorities in February 2003. A case was initially defined by clinical criteria; a suspected or probable case was defined as an illness that included potential exposure to an existing case and fever with pneumonia or respiratory distress syndrome. In April 2003, a confirmed case was defined as a case from which SARS-CoV was isolated from culture.[22] SARS-CoV infections have an incubation period of 2 to 10 days. Systemic symptoms such as fever and chills followed by a dry cough and shortness of breath begin within 2 to 7 days. Patients may develop pneumonia and lymphopenia by days 7 to 10 of the illness. Most patients with SARS-CoV have a clear history of exposure either to a patient with SARS or to a setting in which SARS-CoV is known to exist. Laboratory tests may be helpful but do not reliably detect infection early during the illness. SARS-CoV should be suspected in patients requiring hospitalization for radiographically confirmed pneumonia or acute respiratory distress syndrome of unknown etiology and one of the following risk factors during the 10 days before the onset of illness: (1) travel to China, Hong Kong, or Taiwan, or close contact with an ill person having a history of such travel; (2) employment in an occupation associated with a risk for SARS-CoV exposure; or (3) inclusion in a cluster of cases of atypical pneumonia without an alternative diagnosis.

A "respiratory hygiene/cough etiquette" strategy should be adopted in all SARS-affected health care facilities. All patients admitted to the hospital with suspected pneumonia should receive the following measures: (1) they should be placed in droplet isolation until it is determined that isolation is no longer indicated (standard precautions are appropriate for most community-acquired pneumonias; droplet precautions for nonavian influenza); (2) they should be screened for risk factors of possible exposure to SARS-CoV; and (3) they should be evaluated with a chest radiograph, pulse oximetry, complete blood count, and additional workup as indicated. If the patient has a risk factor for SARS, droplet precautions should be implemented pending an etiologic diagnosis. When there is a high index of suspicion for SARS-CoV disease, the patient should be treated in terms of SARS isolation precautions immediately (including airborne precautions), and all contacts of the ill patient should be identified, evaluated, and monitored.[22] Although ribavirin, high-dose corticosteroids, and interferons have been used in treatment, it is unclear what effect they have had on clinical outcome. No definitive therapy has been established. Empiric antibiotic treatment for community-acquired pneumonia following the current American Thoracic Society/Infectious Diseases Society of America guidelines is recommended pending etiologic diagnosis. Diagnostic tests for SARS-CoV include antibody testing using an enzyme immunoassay and RT-PCR tests for respiratory, blood, and stool specimens.[23] In the absence of known SARS-CoV transmission, testing is recommended only in consultation with public health authorities. Testing for influenza, respiratory syncytial virus, pneumococcus, chlamydia, mycoplasma, and legionella should be conducted, as the identification of one of these agents excludes SARS by case definition. Clinical samples can be obtained during the first week of illness with a nasopharyngeal swab plus an oropharyngeal swab and a serum or a plasma specimen. After the first week of illness, a nasopharyngeal swab plus an oropharyngeal swab and a stool specimen should be obtained. Serum specimens for SARS-CoV antibody testing should be collected when the diagnosis is first suspected and at later times as indicated. An antibody response can occasionally be detected during the first week of illness, is likely to be detected by the end of the second week of illness, and at times may not be detected until more than 28 days after the onset of symptoms. Respiratory specimens from any of several different sources may be collected for viral and bacterial diagnostics, but the preferred specimens of choice are nasopharyngeal washes or aspirates.[23]

Middle East Respiratory Syndrome (MERS-CoV)

MERS-CoV is a disease caused by a coronavirus and results in severe acute respiratory disease including fever, chills, cough, and dyspnea. Some patients also develop nausea, vomiting, and diarrhea. It has a 30% mortality rate. Most patients who have died had underlying comorbidities and developed pneumonia or renal failure. The illness was first reported in Saudi Arabia in September 2012; most cases appear to be limited to the Arabian Peninsula. The incubation period ranges from 2 to14 days. The illness can spread from person to person but it requires close contact, and no sustained transmission had been reported by late 2014. The virus could evolve and lead to sustained transmission, but this cannot be predicted with certainty. There is no vaccine for MERS-CoV and there is no specific treatment, but there is ongoing research by the National Institutes of Health (NIH) and other entities to fill this gap.[24]

Nipah and Hendra Viruses

The Nipah and Hendra viruses are closely related but distinct paramyxoviruses that compose a new genus within the family Paramyxoviridae. The Nipah virus was discovered in Malaysia in 1999 during an outbreak of a zoonotic infection, now called Nipah virus encephalitis, involving mostly pigs and some human cases.[25] Hendra, the causative

agent of Hendra virus disease, was identified in a similar outbreak involving a single infected horse and three human cases in Southern Australia in 1994.[26] It is believed that certain species of fruit bats are the natural hosts for these viruses and remain asymptomatic. Horses and pigs act as amplifying hosts for the Hendra and Nipah viruses, respectively. The mode of transmission from animal to humans appears to require direct contact with tissues or body fluids or with aerosols generated during butchering or culling. Personal protective equipment including gowns, gloves, and respiratory and eye protection is advised for agricultural workers culling infected animal herds. Thus far, human-to-human transmission of these viruses has not been reported.

In symptomatic cases, the onset of disease begins with flu-like symptoms and rapidly progresses to encephalitis with disorientation, delirium, and coma. Fifty percent of those with clinically apparent infections have died from their disease. There is currently no approved treatment for these infections, and, therefore, therapy relies heavily on supportive care. The antiviral drug ribavirin has been used in past infections, but its effectiveness remains unproven in clinically controlled studies.[27] Although no person-to-person transmission is known to have occurred, barrier nursing and droplet precautions are recommended because respiratory secretions and other bodily fluids are known to harbor the virus. The clinical laboratory should be notified before specimens are sent as these may pose a laboratory hazard. Specimens for viral isolation and identification should be forwarded to a reference laboratory. Requests for testing should come through public health departments, which should contact the CDC Emergency Operations Center at 770-488-7100 before sending specimens.

Biological Response Modifiers

BRMs direct the myriad complex interactions of the immune system. BRMs include erythropoietins, interferons, interleukins, colony-stimulating factors, granulocyte and macrophage colony-stimulating factors, stem cell growth factors, monoclonal antibodies, tumor necrosis factor inhibitors, and vaccines.[28]A growing understanding of the structure and function of BRMs is driving the discovery and creation of many novel compounds including synthetic analgesics, antioxidants, and antiviral and antibacterial substances. For example, BRMs are being used to treat debilitating rheumatoid arthritis by targeting cytokines that contribute to the disease process.[29] By neutralizing or eliminating these targeted cytokines, BRMs may reduce symptoms and decrease inflammation. BRMs may also be used as anticarcinogens, with the following goals: (1) to stop, control, or suppress processes that permit cancer growth; (2) to make cancer cells more recognizable, and therefore more susceptible, to destruction by the immune system; (3) to boost the killing power of immune system cells, such as T cells, natural killer cells, and macrophages; (4) to alter growth patterns in cancer cells to promote behavior like that of healthy cells; (5) to block or reverse the processes that change a normal cell or a precancerous cell into a cancerous cell; (6) to enhance the ability of the body to repair or replace normal cells damaged or destroyed by other forms of cancer treatment, such as chemotherapy or radiation; and (7) to prevent cancer cells from spreading to other parts of the body.[30,31]

More of these promising new drugs are currently in development. It can be readily theorized that research to develop various BRMs can be subverted to a malicious end. That is, instead of using BRMs to suppress cancer growth or to decrease disease susceptibility, researchers could develop compounds to cause illness and death. Other drugs could be designed to alter certain metabolic processes or to alter brain chemistry to affect cognition or mood. The opportunity for mischief is limited only by the imagination of the person with ill intent.

Synthetic Biology and Bioengineered Pathogens

The field of synthetic biology had its beginnings near the turn of the millennium with the idea that basic engineering principles could be applied to biological systems at the cellular and genetic levels to create new and improved organisms. Synthetic biology is the "engineering of biology."[32] The goal and end products of the engineering are conventionally to be used for the benefit of humankind. However there has been increasing concern that synthetic biology could be used for nefarious purposes.[33–36]

A precise definition of synthetic biology has not yet been established; however, a consensus is building that synthetic biology is defined as the use of molecular biology tools and techniques to forward the engineering of cellular behavior. There is disagreement among scientists whether the new field of synthetic biology will allow terrorists to create biological agents with more lethal characteristics or create completely novel pathogens with enhanced pathogenicity and weaponization potential in an easier fashion.[37] Some argue that synthetic biology causes "de-skilling" of biological techniques and allows laboratory processes to become easier for less experienced scientists or even laypeople to master.[35] Others maintain that the tacit or unwritten laboratory skills that only a few highly trained scientists possess and which are very difficult to transfer and very difficult for a terrorist to acquire.[38] Social scientists have carefully studied these tacit skills and argue that these techniques are very difficult to pass on from scientist to scientist without considerable effort that is often impossible even under the most ideal circumstances.[38] This difficulty was historically present in both the U.S. and Soviet biological weapons programs each of which were extremely well funded and staffed with competent scientists.

Nevertheless, the rapid advance of synthetic biology has the potential to alter the present and future threat of biological weapons.[9,35,36] Already, complete or partial genomic sequence data for many of the most lethal human pathogens (such as anthrax, plague, and the smallpox virus) have been published and are widely available via the Internet.[39] In addition to the enormous explosion in our knowledge of human pathogens, there is a parallel increased understanding of the complexities of the human immune response to foreign agents and toxins. Such knowledge has led to a deeper understanding of the development of basic immunity to a variety of different human infectious diseases.

With this increase in scientific knowledge has come the power to manipulate the immune system at its most fundamental level. As we prepare for future threats, we must not ignore the potential quantum leap that synthetic biology offers to terrorists for developing new biological-warfare threats. Examples of biological threats that could be produced through the use of synthetic biology include the following: (1) microorganisms resistant to antibiotics, standard vaccines, and therapeutics; (2) innocuous microorganisms genetically altered to produce a toxin, a poisonous substance, or an endogenous bioregulator; (3) microorganisms possessing enhanced aerosol and environmental stability characteristics; (4) immunologically altered microorganisms able to defeat standard threat identification and diagnostic methods; (5) genetic vectors capable of transferring human and foreign genes into human cells for therapeutic purposes[39]; and (6) combinations of these with improved delivery systems.[40–42]

POTENTIAL FUTURE CHEMICAL WEAPONS

Nature of the Problem

The threats associated with the use of chemical weapons as battlefield or terrorist weapons are not easy to assess.[43,44] Risk assessment of use must take into account national laws, international treaties and

conventions, and the likelihood of adherence to these legal obligations. Loopholes in existing agreements can be exploited to develop weapons that are technically not prohibited by international law. Goals and objectives may vary depending on whether military use is planned at the strategic, tactical, or operational level and whether the developer is a national government, a breakaway republic, a kidnapped or recruited scientist, or a terrorist cell. Risk of use may also depend on whether the targets are military versus civilian, human versus nonhuman (animals or plants, including livestock and crops), or individual (as in assassinations) versus large groups, and on whether the aim is death versus incapacitation. Risk also depends on agent availability and on the technology available for production, storage, and dissemination; current advances in technology are associated with a higher risk of weaponization. Two examples from the twentieth century and one from the twenty-first can illustrate the fallibility of intelligence:

1. During most of World War II, the Allied perception of risk from possible chemical agent use by Axis powers focused on those agents, primarily pulmonary agents and vesicants, known from World War I. In fact, Germany had developed a new kind of chemical-warfare agent, the compounds later to be called G-series nerve agents. Their existence came as a complete surprise to Western governments when, in the waning days of the European campaign, Allied soldiers advancing into Germany discovered buried nerve-agent munitions and entire nerve-agent factories. Why these agents were never used on the battlefield is a topic of much speculation, but in retrospect they clearly posed the most lethal, yet unrecognized, threat from Germany.[45]
2. Assessment of the chemical threat posed by Saddam Hussein at the time of the Gulf War of 1991 centered on the known Iraqi use of sulfur mustard and nerve agents during the Iran-Iraq War in the 1980s. It was not until 1998 that Reuters News Agency reported the discovery by British intelligence that Iraq had stockpiled large quantities of a "mental incapacitant" (incapacitating agent) known as Agent 15.[18,46]
3. The risk of use of chemical agents by Iraq after 2001 was assessed to be high partly because of the known stockpiles of sulfur mustard and nerve agents (as well as the suspected stockpiles of cyanide and the new revelations about Agent 15) from the time of the 1991 Gulf War. President George W. Bush's summary in October of 2002 of the National Intelligence Estimates (NIE) states, "Baghdad has begun renewed production of mustard, sarin, GF (cyclosarin), and VX. Although information is limited, Saddam probably has stocked at least 100 and possibly as much as 500 metric tons of CW agents." However, in 2006 the Iraq Study Group (ISG) Report was released and reported that "while a small number of old, abandoned chemical munitions have been discovered, ISG judges that Iraq unilaterally destroyed its undeclared chemical weapons stockpile in 1991."[47] Although the initial intelligence proved to be wrong, it does not invalidate the argument that the risk from these agents, if possessed, would be very concerning.

Chemical agents originally used during World War I are sometimes considered obsolete, especially in comparison to the more potent nerve agents and incapacitating agents. However, agent potency is only one part of the story. To deliver the 5 μg that represents an estimated lethal dose for half of an exposed group (LD_{50}) of the nerve agent VX would seem to be easier than delivering the 3 to 7 g that constitute the LD_{50} of sulfur mustard and more difficult than delivering the much smaller lethal doses of toxins such as botulinum toxin.[48] In fact, sulfur mustard is easier to synthesize than is a nerve agent and is easy to disseminate in a clandestine manner to create delayed effects. Thus mustard still lays claim to being the "King of Gases," and it has allegedly been used in a variety of venues since the end of World War II. Most known chemicals with toxicities equal to or greater than that of ammonia could theoretically be used as chemical warfare or terrorism agents.

Existing Agents and Their Potential for Future Use

Existing chemicals capable of weaponization for military or terrorist use include the following:

1. Battlefield and riot-control agents
   a. Pulmonary agents (see Chapter 114)
   b. Vesicants (see Chapter 113)
   c. Cyanide (see Chapter 115)
   d. Nerve agents (see Chapter 112)
   e. Antimuscarinic agents such as BZ and Agent 15 (see Chapter 116)
   f. Riot-control agents (see Chapter 120)
   g. Defoliants and other herbicides
   h. Novichok
2. New chemicals employed for physicochemical effects
   a. Related compounds
   b. Battlefield incendiary agents, smokes (including standard military white obscurant smoke, or HC smoke), and other combustion products such as oxides of nitrogen and perfluoroisobutylene (PFIB)
   c. Opioids (see Chapter 118) and other anesthetic agents (see Chapter 122)
   d. Cholinergic agents (see Chapter 121)
   e. Psychedelic indoles and other hallucinogens (see Chapter 117)
3. Toxic industrial chemicals or materials (see Chapter 111)
4. Poisons
5. Combination of chemicals
6. Nontraditional agents (see Chapter 119) such as hydrofluoroalkane propellant attack

Existing chemicals remain candidate agents for future use. Some compounds not developed to cause injury or incapacitation nevertheless can be very dangerous; hexachloroethane (HC) smoke, for example, can cause the same type of pulmonary damage induced by phosgene, a chemical weapon used in World War I. The CDC lists nearly 70 separate chemicals, including a variety of toxic industrial chemicals and poisons, as potential agents for terrorism. These include osmium tetroxide, long-acting anticoagulants, heavy metals, toxic alcohols, and white phosphorus.[49] The April 21, 2000, *Morbidity and Mortality Report* included an even longer list of chemical agents that might be used by terrorists.[50] Pyrolysis, the thermochemical change of an organic material in the absence of oxygen by heat, and products from explosions and conflagrations may release large quantities of cyanide and other toxicants that, although different from the original chemicals present, may still cause death. Industrial chemicals are readily available in large quantities as preformed compounds and should be considered high on the list of potential terrorist agents.[51,52] Toxins that are chemicals produced within biological organisms also represent high-threat agents.[53] New chemicals are currently being synthesized on rigid three-dimensional molecular skeletons, the most promising of which are the norbornanes. Norbornane is a bicyclic crystalline hydrocarbon (C_7H_{12}).[54] Building on norbornane geometry allows for a modular enhancement of the number of functional sites on a given molecule. Many norbornane derivatives, such as the mixture of chlorobornanes known as the toxaphenes, are persistent and have significant acute and chronic toxicity. These norbornane derivatives have been considered as potential candidates for new agents. Novichok [55–58] (Russian for "newcomer") refers to the alleged Russian development of a highly toxic binary nerve agent or generation of nerve agents (sometimes called "fourth-generation" agents). Only sketchy and unverifiable information is available in the unclassified literature, but the existence of these agents would demonstrate the possibility of creating new

chemical compounds toxic enough to be used as chemical warfare or terrorist agents. One of the sources of unclassified information is from a dissident Russian scientist who wrote newspaper articles and published a book about the Novichok program and the types of chemical agents that were produced.[59] So-called GV analogs combining some of the properties of G-series and V-series nerve agents have also been suggested as potential new agents.[38]

The use in 2002 of an incapacitating gas in the siege of a Moscow theater taken over by Chechen rebels was evidence of use of a chemical aerosol.[60,61] The Russian Health Minister at the time, after significant international pressure identified the aerosol as a fentanyl derivative and then stated that use of a fentanyl derivative was not prohibited by the Chemical Weapons Convention. Further investigations of survivors have suggested that carfentanil and remifentanil were possibly used in the siege.[62]

Organofluorines have been investigated because of their reported ability to defeat protective-mask or chemical-filtration systems.[38] Other incapacitating agents under development exert primarily physical rather than chemical effects and include immobilizing agents ("stickums"), antitraction gels ("slickums"), and malodorants.[63,64] An effective incapacitating agent must be highly potent and reversible. It also must have rapid onset, short duration of action, and a high safety margin.[63]

Nontraditional agents (NTAs) are chemicals that do not fall in the traditional chemical weapons category but have been reportedly researched or developed for use as chemical weapons. "NTAs are novel chemical threat agents or toxicants requiring adapted countermeasures," according to Homeland Security Presidential Directive/HSPD-18.[65] Developing defenses against NTAs is a listed priority for the U.S. Department of Defense.[41]

Technological Modifications of Battlefield Chemical Agents and Delivery Systems

Ways in which existing or future battlefield chemical agents and delivery systems could be modified to improve performance must be considered. These modifications include the following:

1. Agent thickening
2. Binarization
3. Micronization: "dusty agents"
4. Developments in delivery systems
   a. Dual-use cyberinsects and biorobots
   b. Nanotechnology

Small quantities of thickening agents, such as acrylates, can be added to chemical agents to increase their viscosity. Thickened agents are more persistent in the environment and in wounds than are nonthickened agents, and they are less easily decontaminated.[66] Although no nation is currently known to stockpile thickened agents, the technology for their production is relatively simple and requires only standard chemical-warfare agents and the right proportion of a thickener.[67] Many industrial chemicals and other poisons could theoretically be rendered more effective as battlefield or terrorist agents by thickening.

In the 1950s, the U.S. Army began to investigate the then-new technology of binarization, although production did not accelerate until the 1960s and deployment was not widespread until the 1980s.[58] A binary chemical weapon did not employ a new kind of agent but rather represented a novel way of producing and storing an already existing type of agent. The idea was to make storage of chemical rounds safer by stopping the production process at the penultimate synthetic step, resulting in two precursor compounds that when mixed would create the desired agent. These two precursors could then be stored separately. Just before use, one component could be inserted into a round, where it would be separated from the other precursor by a thin membrane. The impact and momentum of the launch of the projectile would burst the membrane to allow for mixing of the components and in-flight production of the chemical agent. In practice, this process was often not complete, but the 20% or so of ancillary reaction product was often extremely toxic by itself. Binarization or some similar production-arrest method could theoretically be used by a clandestine terrorist cell to help evade detection and to decrease the risks associated with the production, transportation, and use of chemical agents.

Micronization is a type of particularization involving the production of extremely fine particles onto which a chemical agent can be adsorbed. During World War II, Germany explored particularization of sulfur mustard onto small carrier particles of silica (silicon dioxide), although other powdered silicates (e.g., talc, diatomite, and pumice) and clays (e.g., kaolinite and Fuller's earth) can also be used.[68] The advantages of such "dusty agents" are increased volatility, facilitation of the movement of relatively nonvolatile agents such as sulfur mustard and the persistent nerve agent VX into the alveoli, and increased penetration of clothing and chemical protective equipment.[31] Iraq used a "dusty mustard" composed of 65% sulfur mustard adsorbed onto silica particles ranging in diameter from 0.1 to 10 μm during its war with Iran. Micronization of a variety of chemical, biological, and toxin agents requires a certain degree of technological sophistication that is becoming increasingly easy to acquire.

Agent delivery can potentially be modified in a variety of ways in addition to thickening and micronization. The Jordanian government released a report in 2004 of the discovery of an elaborate plot by Al Qaeda terrorists for a two-stage attack using a massive vehicle-borne improvised explosive device followed by the release of toxic chemicals to include acetones, nitric acid, and sulfuric acid.[69] Similarly, enhanced-fragmentation munitions could be used in combination with chemical agents to drive the agents more effectively into the body.

Innovative new delivery systems taking advantage of advances in robotics include the proposed use of cyberinsects and biorobots to deliver biological agents, chemical agents, or toxins.[70] Engineering on an even smaller scale is the purview of nanotechnology, also called "micromechanical engineering" and "micro-electromechanical systems."[71] Nanotechnology takes advantage of the unique properties of materials on the scale of about a nanometer (10 to 9 m) 72 and deals with the molecule-by-molecule or even atom-by-atom assembly of materials. Nanoparticles behave in unusual and unpredictable ways, are small enough to enter cells easily, and in fact are being developed to provide not only better storage and dispersal of pharmaceutical products but also more efficient transport of both biological organisms (e.g., viruses) and chemical compounds into the body.[71] In some cases they may be surprisingly toxic, partly because of the ease with which they can cross membranes, including the blood-brain barrier, and enter cells.[73] This toxicity could be exploited by governments or terrorist organizations interested not only in small-particle delivery of chemical agents but also in the ancillary and perhaps synergistic effects of the carrier materials themselves.

Nanomaterials can be encapsulation compounds such as fullerenes, or buckyballs, which are hollow 60-carbon geodesic shells; nanoshells (e.g., a gold shell surrounding an inert silica core); a "self-assembled, polyamino acid nanoparticles system" under development in France; or dendrimers, which are onion-like layers of shells surrounding a biologically active core.[72] Any of these materials could be used to deliver existing or new chemical agents. Other nanomaterials include self-assembling liquids composed of cylindrical nanofibers (each 6 to 8 nm in diameter) that solidify upon injection to form structured scaffolds capable of presenting ordered peptide signals to cells. A ferrofluid such as a colloidal suspension of nanoscale ferrous oxide can be coupled

with antibodies in a laboratory to detect and concentrate rare human cells in a diagnostic setting, but this technology could easily be adapted to target those cells in vivo.

Quantum dots are nanoscale semiconductor crystals that show promise in the in vitro and in vivo diagnosis of a variety of conditions; although their main use is projected to be in the laboratory, animal experimentation involving injected quantum dots has demonstrated successful targeting of lymph nodes and of prostate-cancer xenografts in mice.

Adverse health effects from any of these kinds of nanoparticles could represent a primary goal for military or terrorist operatives in addition to the toxicity of any other chemicals delivered by the nanoparticles. For example, water-soluble fullerenes (or buckyballs) have caused brain damage in largemouth bass.[74] Also, dendrimers can cause osmotic and membrane damage and can activate the clotting and complement systems. Quantum dots composed of selenium, lead, and cadmium could release those metals into cells, depending on the composition of the surface coating of the dots, and cause damage.[72]

"Designer" Chemicals from Biotechnological Processes

Biotechnology refers to "any technological application that uses biological systems, living organisms, or derivatives thereof, to make or modify products or processes for specific use."[75] Biotechnology includes such time-honored practices as the baking of bread and the brewing of beer, but in the twenty-first century refers in particular to genetic engineering, that is, the artificial transfer of genes from one organism to another and the consequent alteration of the genetic structure of a cell.[76] It is founded on the basic sciences of genomics (the study of the genetic composition of an organism) and proteomics (the study of the expression of the genome by means of protein synthesis). "Designer" chemicals could be produced from biotechnological processes. These processes include the following: (1) combinatorial chemistry and ligand modification; (2) genomics and target identification; (3) microarrays, proteomics, and rational agent design; and (4) toxicogenomics, database mining, and the prediction of toxicity.[77] These developments, if used for chemical warfare agents, would be considered "dual-use technology." This is technology that can be used for both peaceful and military aims.

Combinatorial chemistry is the production of complex sets, or so-called libraries, of related compounds, as in the case of the norbornane derivatives previously described. Automated screening techniques to select for library elements with desired toxic effects on specified target organs can process several hundred thousand compounds a day against several dozen different proteins. This obviously accelerates tremendously the development of new chemical agents.

Genomics has benefited enormously from three modern scientific efforts: the Human Genome Project, the Human Genome Diversity Project, and gene therapy.[78] Identification and cataloging of hundreds of single-nucleotide polymorphisms (individual sequence variations) allow for the selection of genomic sequences to be mass-produced for insertion into cells to create a specific effect. Targeting unusual sequences of high prevalence in certain populations raises the specter of genomic, or ethnic, weapons, as previously described. Less appreciated is the potential for genomics to be used to develop drugs and chemical or toxin agents that can also be targeted to specific variants within a population of humans, animals, or crops. The widespread availability of genome libraries on the Internet makes it nearly impossible to control or restrict access to the already published genomic libraries on over a hundred microbial pathogens.[79]

Proteomics complements genomics by characterizing the protein expression of segments of the genome and by making it easier to develop compounds that target or produce a specific protein. Direct gene insertion, genetic delivery via virus or bacteria, or drug tailoring to affect a given protein can be used. For example, a scorpion toxin has been successfully engineered into a virus that acts as a pesticide against caterpillars. Protein sequences in toxins are partly responsible for resistance to light, oxygen, moisture, and desiccation; the insertion of genes to create altered proteins or the introduction of chemical agents engineered to cause structural changes in expressed proteins could significantly alter the toxicity of a given compound.[77] Furthermore, the widespread use of DNA microarrays (glass slides or chips imprinted with thousands of specific single-stranded DNA sequences) allows for fast-automated screening of candidate compounds.

Scientists involved in the selection and evaluation of specific chemical agents can now use toxicogenomics (the study of genetic variation of response to toxins) and data mining (the computerized analysis of databases of drug and chemical information via sophisticated neural nets) as tools to eliminate less likely candidates and to algorithmically predict compounds with high toxicity or with other desired characteristics relating to environmental persistence, toxicokinetics (absorption, distribution, biotransformation, and elimination), and toxicodynamics (mechanism of action). Such tools will undoubtedly lead to the development not only of new pharmaceutical agents but also of designer toxins for military or terrorist use.[77]

CONCLUSIONS

If history is any guide, new biological and chemical weapons and novel "mid-spectrum" agents (e.g., toxins, bioregulators, synthetic viruses, and genocidal weapons) will be developed in the future, and new modifications will be found to improve the production, weaponization, storage, delivery, and action of existing agents.[33,80–82] Naturally occurring emerging infectious diseases provide examples of newly identified pathogens with weaponization potential, and midspectrum agents such as toxins and bioregulators will undoubtedly assume more prominence with the accelerating pace of synthetic biology. Agents of any category can theoretically be engineered to target specific genes or proteins with differential population prevalence to produce genomic or ethnic weapons; and advances in proteomics, toxicogenomics, and computerized database mining could be used for the rapid and efficient development of not only new drugs but also new chemical agents for terrorism.[9,17,68] Synthetic biology has now advanced to the point that no special equipment is required beyond that available to any modern molecular-biology laboratory, and the scale of operations is also well within the means of governments and terrorist groups.[78] The threats from future modification of existing agents and from the development of new agents, new agent-development technologies, and innovative delivery systems should not and must not be underestimated.

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REFERENCES

1. Joy RJT. Historical aspects of medical defense against chemical warfare. In: Sidell FR, Takafuji ET, Franz DR, eds. *Medical Aspects of Chemical and Biological Warfare, Part I.* Washington, DC: Borden Institute; 1997:111–128.
2. Mann J. *Murder, Magic, and Medicine. Oxford.* New York, NY: Oxford University Press; 1992.
3. Fenn EA. *Pox Americana: The Great Smallpox Epidemic of 1775-82.* Hill and Wang: New York, NY; 2001.
4. Williams PWD. *Unit 731: Japan's Secret Biological Warfare in World War II.* London: Hodder and Stoughton; 1989.

5. Christopher GW, Cieslak TJ, Pavlin JA, Eitzen EM, Jr. Biological warfare. A historical perspective. *JAMA*. 1997;278(5):412–417.
6. Regis E. *The Biology of Doom: America's Secret Germ Warfare Project*. Henry Holt: New York, NY; 1999.
7. Meselson M, Guillemin J, Hugh-Jones M, et al. The Sverdlovsk anthrax outbreak of 1979. *Science*. 1994;266(5188):1202–1208.
8. Noah DL, Huebner KD, Darling RG, Waeckerle JF. The history and threat of biological warfare and terrorism. *Emerg Med Clin North Am*. May 2002; 20(2):255–271.
9. Tucker JB. *Toxic Terror: Assessing Terrorist Use of Chemical and Biological Weapons*. Cambridge, MA: MIT Press; 2000.
10. CDC. Follow-up of deaths among U.S. Postal Service workers potentially exposed to *Bacillus anthracis*–District of Columbia, 2001-2002. *MMWR Morb Mortal Wkly Rep*. 2003;52(39):937–938.
11. Fennelly KP, Davidow AL, Miller SL, Connell N, Ellner JJ. Airborne infection with *Bacillus anthracis*—from mills to mail. *Emerg Infect Dis*. 2004;10(6):996–1002.
12. UN Secretary General. United Nations Mission to Investigate Allegations of the Use of Chemical Weapons in the Syrian Arab Republic Report on the Alleged Use of Chemical Weapons in the Ghouta Area of Damascus on 21 August 2013. 2013.
13. Alibek KHS. *Biohazard : The Chilling True Story of the Largest Covert Biological Weapons Program in the World, Told from the Inside by the Man Who Ran It*. New York, NY: Random House; 1999.
14. Morens DM, Folkers GK, Fauci AS. The challenge of emerging and re-emerging infectious diseases. *Nature*. 2004;430(6996):242–249.
15. Phillips HKD. *The Spanish Influenza Pandemic of 1918-19: New Perspectives*. London; New York, NY: Routledge; 2003.
16. Hummel S. Ebola: not an effective biological weapon for terrorists. *CTC Sentinel*. 2014;7(9):16–19.
17. Centers for Disease Control and Prevention. *Emergency Preparedness and Response: Bioterrorism Agents/Diseases*. http://emergency.cdc.gov/agent/agentlist-category.asp. Accessed November 23, 2014.
18. Centers for Disease Control and Prevention. *Avian Influenza A Virus Infections of Humans*. 2008. http://www.cdc.gov/flu/avian/gen-info/avian-flu-humans.htm.
19. Stevens J, Corper AL, Basler CF, Taubenberger JK, Palese P, Wilson IA. Structure of the uncleaved human H1 hemagglutinin from the extinct 1918 influenza virus. *Science*. 2004;303(5665):1866–1870.
20. Centers for Disease Control and Prevention. Interim Guidance for Protection of Persons Involved in U.S. Avian Influenza Outbreak Disease Control and Eradication Activities. 2006. Available at: http://www.cdc.gov/flu/avian/professional/protect-guid.htm. Accessed April 11, 2015.
21. Teleman MD, Boudville IC, Heng BH, Zhu D, Leo YS. Factors associated with transmission of severe acute respiratory syndrome among health-care workers in Singapore. *Epidemiol Infect*. 2004;132(5):797–803.
22. CDC. Revised U.S. surveillance case definition for severe acute respiratory syndrome (SARS) and update on SARS cases—United States and worldwide, December 2003. *MMWR Morb Mortal Wkly Rep*. 2003;52 (49):1202–1206.
23. Centers for Disease Control and Prevention. *Severe Acute Respiratory Syndrome: Public Health Guidance for Community-Level Preparedness and Response to SARS*. Version 2. Supplement F: Laboratory guidance. Appendix F8—Guidelines for laboratory diagnosis of SARS-CoV infection. 2005. Available at: http://www.cdc.gov/sars/guidance/. Accessed April 11, 2015.
24. Centers for Disease Control and Prevention. *About MERS*. Available at: http://www.cdc.gov/coronavirus/mers/. Accessed November 23, 2014.
25. Prevention CfDCa. *Hendra Virus Disease & Nipah Virus Encephalitis*. Available at: http://www.cdc.gov/ncidod/dvrd/spb/mnpages/dispages/Fact_Sheets/Hendra_Nipah_Fact_Sheet.pdf. Accessed April 11, 2015.
26. Mackenzie JS. Emerging viral diseases: an Australian perspective. *Emerg Infect Dis*. 1999;5(1):1–8.
27. Snell NJ. Ribavirin therapy for Nipah virus infection. *J Virol*. 2004;78 (18):10211.
28. Kagan E. Bioregulators as instruments of terror. *Clin Lab Med*. Sep 2001;21 (3):607–618.
29. O'Dell JR. Therapeutic strategies for rheumatoid arthritis. *N Engl J Med*. 2004;350(25):2591–2602.
30. Bokan S, Breen JG, Orehovec Z. An evaluation of bioregulators as terrorism and warfare agents. *ASA Newslett*. Available at: http://www.asanltr.com/newsletter/02-3/articles/023c.htm. Accessed April 11, 2015.
31. U.S. Congress, Office of Technology Assessment. *Technologies Underlying Weapons of Mass Destruction*. US GPO: Washington, DC; 1993.
32. Cameron DE, Bashor CJ, Collins JJ. A brief history of synthetic biology. *Nat Rev Microbiol*. 2014;12(5):381–390.
33. Casadevall A. The future of biological warfare. *Microb Biotech*. 2012;5 (5):584–587.
34. Garrett L. Biology's brave new world: the promise and perils of the synbio revolution. *Foreign Affairs* 2013. Available at: http://www.foreignaffairs.com/articles/140156/laurie-garrett/biologys-brave-new-world. Accessed October 29, 2014.
35. Tucker JB. Could terrorists exploit synthetic biology? *New Atlantis*. 2011;31:69–81.
36. Tucker JB, Zilinskas RA. The promise and perils of synthetic biology. *New Atlantis*. 2006;12(1):25–45.
37. Jefferson C, Lentzos F, Marris C. *Synthetic Biology and Biosecurity: How Scared Should We Be*. London: King's College London; 2014.
38. Ouagrham-Gormley SB, Vogel KM. The social context shaping bioweapons (non)proliferation. *Biosecur Bioterr*. 2010;8(1):9–24.
39. Black JL, 3rd. Genome projects and gene therapy: gateways to next generation biological weapons. *Mil Med*. 2003;168(11):864–871.
40. Appel JM. Is all fair in biological warfare? The controversy over genetically engineered biological weapons. *J Med Ethics*. 2009;35(7):429–432.
41. Program DoDCaBD. *2013 Department of Defense Chemical and Biological Defense Annual Report to Congress;* 2013.
42. Roberts MA, Cranenburgh RM, Stevens MP, Oyston PC. Synthetic biology: biology by design. *Microbiol*. 2013;159(Pt 7):1219–1220.
43. Cordesman AH. *Defending America: Iraq and other threats to the US involving weapons of mass destruction;* 2001. Available at: http://csis.org/files/media/csis/pubs/iraq_otherthreatswmd.pdf, Accessed October 15, 2014.
44. U.S. Government Accountability Office. *Combating Terrorism: Need for Comprehensive Threat and Risk Assessments of Chemical and Biological Attacks*. Report to Congressional requesters. Washington, DC. 1999. GAO/NSIAD-99-163.
45. Sidell FR. Nerve agents. In: Sidell FR, Takafuji ET, Franz DR, et al., eds. *Medical Aspects of Chemical and Biological Warfare, Part I*. Washington, DC: Borden Institute; 1997:111–128.
46. Fitzgerald GM, Sole DP. *CBRNE: incapacitating agents, Agent 15;* 2014. Available at: http://emedicine.medscape.com/article/833238-overview, Accessed October 15, 2014.
47. National Intelligence Estimate: Iraq's Continuing Programs for Weapons of Mass Destruction. In: Intelligence DoC, Washington, DC, 2002. Available at: https://www2.gwu.edu/~nsarchiv/NSAEBB/NSAEBB129/nie.pdf; accessed April, 11, 2015.
48. Pitschmann V. Overall view of chemical and biochemical weapons. *Toxins*. 2014;6(6):1761–1784.
49. Centers for Disease Control and Prevention. *Chemical Agents*. 2013. Available at: http://emergency.cdc.gov/agent/agentlistchem.asp. Accessed October 15, 2014.
50. CDC. Biological and chemical terrorism: strategic plan for preparedness and response. Recommendations of the CDC Strategic Planning Workgroup. *MMWR Recomm Rep*. 2000;49(Rr-4):1–14.
51. Burklow TR, Yu CE, Madsen JM. Industrial chemicals: terrorist weapons of opportunity. *Pediatr Ann*. 2003;32(4):230–234.
52. U.S. Department of Health Human Services. Agency for Toxic Substances Disease Registry. Industrial chemicals and terrorism: human health threat analysis, mitigation and prevention. Available at: http://www.mapcruzin.com/scruztri/docs/cep1118992.htm. Accessed April 11, 2015.
53. Madsen JM. Toxins as weapons of mass destruction. A comparison and contrast with biological-warfare and chemical-warfare agents. *Clin Lab Med*. Sep 2001;21(3):593–605.
54. Holderna-Natkaniec K, Natkaniec I, Khavryutchenko V. Neutron spectroscopy of norbornane. *Phase Trans*. 2003;76(3):275–279.
55. Adams JR. Russia's toxic threat. *Wall St J (Midwest Ed)*. 1996;A18.
56. Englund W. Ex-Soviet scientist say Gorbachev's regime created new nerve gas in '91. *Baltimore Sun*. 1992;3A.

57. Sands A, Pate J. CWC compliance issues. In: Tucker JB, ed. *The Chemical Weapons Convention: Implementation Challenges and Solutions*. Washington, DC: Center for Nonproliferation Studies, Monterey Institute of International Studies; 2001:17–22.
58. Smart JK. History of chemical and biological warfare: an American perspective. In: Sidell FR, Takafuji ET, Franz DR, et al., eds. *Medical Aspects of Chemical and Biological Warfare, Part I*. Washington, DC: Borden Institute; 1997:9–86.
59. Mirzayanov V. *State Secrets: An Insider's Chronicle of the Russian Chemical Weapons Program*. Parker, CO: Outskirts Press; 2008.
60. Bismuth C, Borron SW, Baud FJ, Barriot P. Chemical weapons: documented use and compounds on the horizon. *Toxicol Lett*. 2004;149 (1–3):11–18.
61. Coupland RM. Incapacitating chemical weapons: a year after the Moscow theatre siege. *Lancet*. 2003;362(9393):1346.
62. Riches JR, Read RW, Black RM, Cooper NJ, Timperley CM. Analysis of clothing and urine from Moscow theatre siege casualties reveals carfentanil and remifentanil use. *J Anal Toxicol*. 2012;36(9):647–656.
63. Pearson A. Incapacitating biochemical weapons. *Nonproliferation Rev*. 2006;13(2):151–188.
64. Harigel GG. *Introduction to Chemical and Biological Weapons*. Washington, DC: Carnegie Endowment for International Peace; 2001. *http://carnegieendowment.org/2001/01/18/introduction-to-chemical-and-biological-weapons*. Accessed April 11, 2015.
65. Department of Homeland Security. Homeland Security Presidential Directive/HSPD-18. *Medical Countermeasures Against Weapons of Mass Destruction* January 31, 2007.
66. Hurst CG. Decontamination. In: Sidell FR, Takafuji ET, Franz DR, et al., eds. *Medical Aspects of Chemical and Biological Warfare, Part I*. Washington, DC: Borden Institute; 1997:351–359.
67. Takafuji ET, Kok AB. The chemical warfare threat and the military healthcare provider. In: Sidell FR, Takafuji ET, Franz DR, et al., eds. *Medical Aspects of Chemical and Biological Warfare, Part I*. Washington, DC: Borden Institute; 1997:111–128.
68. Dando MR. *The danger to the Chemical Weapons Convention from incapacitating chemicals. First CWC Review Conference Paper No. 4. Strengthening the Chemical Weapons Convention*. Bradford, UK: University of Bradford Department of Peace Studies; 2003.
69. Jordan "was chemical bomb target." 2014. Available at: http://news.bbc.co.uk/2/hi/3635381.stm. Accessed April 11, 2015.
70. DaSilva EJ. Biological warfare, bioterrorism, biodefence and the biological and toxin weapons convention. *Electronic J Biotech*. 1999;2(3):9–128.
71. Jane's Chem-Bio Web. News: Nanotechnology: the potential for new WMD. Available at: http://chembio.janes.com [subscription required]. Accessed April 11, 2015.
72. Perkel J. The ups and downs of nanobiotech. *Scientist*. 2004;18(16).
73. Weiss R. Nanotechnology precaution is urged: miniscule particles in cosmetics may pose risk, British scientists say. *Washington Post*. July 30, 2004, A02.
74. Oberdorster E. Manufactured nanomaterials (fullerenes, C60) induce oxidative stress in the brain of juvenile largemouth bass. *Environ Health Perspect*. 2004;112(10):1058–1062.
75. Wikipedia, The Free Encyclopedia. *Biotechnology*. Available at: http://en.wikipedia.org/wiki/Biotechnology. Accessed April 11, 2015.
76. Convention on Biological Diversity, United Nations Environment Program. Governments to advance work on Cartagena Protocol on Biosafety [press release]. Available at: http://www.biodiv.org/doc/meetings/bs/iccp-03/other/iccp-03-pr-en.pdf. Accessed April 11, 2015.
77. Wheelis M. Biotechnology and biochemical weapons. *Nonproliferation Re*. 2002;9(1):48–53.
78. Barnaby W. *The Plague Makers: The Secret World of Biological Warfare*. 3rd ed. Continuum International: New York, NY; 2000.
79. Russo E. NRC wants genome data unfettered. *Scientist*. 2004. http://www.the-scientist.com/?articles.view/articleNo/23067/title/NRC-wants-genome-data-unfettered/. Accessed October 15, 2014.
80. Aas P. The threat of mid-spectrum chemical warfare agents. *Prehosp Disaster Med*. 2003;18(4):306–312.
81. Caves JP, Carus WS. *The Future of Weapons of Mass Destruction: Their Nature and Role in 2030*. Washington, DC: National Defense University Press; 2014, Occasional Paper 10.
82. Kagan E. Bioregulators as prototypic nontraditional threat agents. *Clin Lab Med*. 2006;26(2):421–443.