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Biological Warfare, Bioterrorism, and Biodefense: An Overview

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Introduction

There is growing concern about the potential use of biological agents (BAs) in war or acts of terrorism accompanied an increased realization that the rapid preparedness and response are needed to prevent or treat the human damage that can be caused by these agents. The use of BAs as weapons, even on a small scale, has the potential for huge social and economic disruption and massive diversion of regional and national resources to combat the threat, to treat primary disease, and to clean up environmental contamination.

History

Biological warfare (BW) is not a twentieth century development. It is certain that ancient man used BW long before recorded history. The use of biological toxins extracted from plants and animals on arrow heads or poison darts to kill game and human enemies certainly predates recorded history. Within recorded history, cases of humans using BW against their fellow humans are well documented from attacking Kafffa with the bodies of plague-victims by Tatars as early as 1346, through the Japanese horrific experiments on human beings at 731 unit in 1932 and attacking at least 11 Chinese cities with the agents of anthrax, cholera, shigellosis, salmonella, and plague, to Iraq's intent to weaponize anthrax, botulinum toxin, and aflatoxin by 1991.

We have seen terrorism emerge as one of the thorniest problems of the post-cold war era. In 1978, Bulgarian dissident Georgi Markov was assassinated using an umbrella gun that shot ricin into his thigh. This terror event would be recorded as a first bioterrorism. Afterwards, many cases of bioterrorism have been reported and analyzed. However, the Monterey Database of incidents involving BAs from 1960 to 1999 indicates that there were few cases in which criminals or terrorists sought to inflict mass casualties with biological agents.

In 1984, *Salmonella typhimurium* bacteria was used to contaminate restaurant salad bars by Rajneeshee cult in The Dalles, Oregon. This event caused 751 cases of food poisoning, but none fatal. The Aum Shinrikyo cult, that released sarin nerve gas in a Japanese subway in 1995, was found to attempt to produce and disperse botulinum toxin and *Bacillus anthracis*. Recently, the anthrax letter attacks on American soil in 2001 have led to a media firestorm of speculation and confusion.

Recent ten years have witnessed a resurgence of concern regarding BW and bioterrorism. In addition to three incidents above, this has occurred for three reasons. The first was the disclosure of a massive Soviet BW program in the 1970s and 1980s, in gross violation of the Biological and Toxin Weapons Convention (BWC). The second was the disclosure in 1995 by the UN Special Commission that, by the time of the Gulf War in 1990, Iraq had prepared a significant BW arsenal and prepared it for battlefield use, essentially without the world's knowledge. The final reason is new and reemerging pathogens such as malaria, shiegellosis, hantavirus, foodborne salmonellosis, plague in India, Ebola hemorrhagic fever, avian (H5N1) influenza, waterborne cryptosporidiosis, AIDS, and suddenly prevailing SARS. Bioterrorism scenarios illustrate the diversity of disciplines and perspectives required to confront these threats.

Characteristics of Biolocial Agents and Bioterrorism

Biological weapons are one of weapons of mass destruction (or mass casualty weapons, to be precise, since they do not damage non-living entities) that are based on bacteria, viruses, rickettsia, fungi, or toxins produced by these organisms. Compared to other types of weapons (nuclear, chemical or conventional), biological weapons are unique in their diversity and capable of 1) causing detrimental changes in the environment, 2) harming or damaging food, water or equipment supplies, 3) causing disease in humans, animals or plants or other living organisms.

The AG developed control lists of dual-use and biological-related materials that are particularly suited for use in BW. These lists currently contain 93 human, animal, and plant biological pathogens and toxins, and dual-use biological-related production equipment. The CDC has identified and classified over thirty of these biological threats in three categories, based on the severity of the threat as well as the seriousness of the diseases that could be caused by their use in bioterrorist or biowarfare acts. BAs categorized into Group A are as follows: Variola virus(smallpox), *Bacillus anthracis*, *Yersinia pestis*, botulinum toxin, *Francisella tularensis,* and filovirus/arenavirus.

BAs are also characterized by the following: Their target system, the nature of the biological agents, whether it is a natural product(random) or one that has been produced by genetic engineering (non-random), technolgy levels needed for weaponization, and their lethal effect.

Biological weapons are easy and cheap to produce and can be used to selectively target humans, animals, or plants. BW agents can cause large numbers of casualties with minimal logistical requirements (in wide area). Perpetrators can escape long before BW agents cause casualties, due to the incubation periods of the agents. For these reasons, BAs have been often called a "Poor Man's Atomic Bomb" or "A Poor Nation's Weapon of Mass Destruction."

Small quantities of lethal BAs can be easily concealed, transported, and released silently into susceptible populations without immediate effects. The spread of disease cannot be controlled until there is awareness of the signs of infection followed by identification of the agent; and if the organism is easily spread from person to person, as in the case of smallpox, the number of casualties could run into the tens of thousands. This fact contributes to both military and civilian vulnerabilities to biological weapons and to the difficulty of providing adequate protection. Because BAs are living

micro-organisms, they shows advantages and disadvantage for weaponizations.

Biological attacks against large populations would most likely be disseminated by aerosol. A respiratory portal of entry may cause different clinical features than naturally occurring disease. Biological attacks could be attempted by contaminating food and water supplies, although modern water purification and the dilution effects in large volumes of water would negate the effectiveness of a water-borne attack.

People don't realize BAs could be the most sophisticated weapons. Because biological weapons could be used covertly, there can be a lot of different deployment scenarios. A lot of different agents could be used in biological weapons. And, there are a lot of different techniques to manufacture biological weapons. Terrorist acts that make use of BAs differ in a number of ways from those involving chemicals.

The BWC of 1972 is an important international agreement aimed at reducing the threat of BW. Unlike the case with the CWC, however, there is no accurate verification mechanism for ensuring compliance with the BWC until now. The diffusion of dual-use technologies relevant to the production of biological and toxin agents, and the potential availability of scientists and engineers formerly employed in sophisticated BW programs such as those of the Soviet Union and South Africa, suggest that the technical barriers to mass-casualty terrorism are eroding. At least seventeen countries are known or suspected of having offensive biological weapons programs, and several of the 30 designated terrorist organizations and other non-state actors, including the Al Qaeda network, have expressed interest in these weapons.

Like any new technologies, biotechnology is a two sided coin. The "good side" allows for the alleviation of human suffering. But there is also a "bad side" that involves biotechnology being used to actually cause human suffering. For example, eugenics, or the science of "improving" a race through genetics, could be greatly enhanced through unethical use of biotechnological practices, i.e., the ethnic bomb. Another possibility of the production and development of biological weapons through biotechnology is so-called the "superbugs", such as an antibiotic-resistant anthrax and plague-ebola hybrids which Soviet researchers tried to create by using genetic engineering in the 1980s. The proliferation of biological weapons is expected to continue, and these weapons could well be used in a regional conflict or terrorist attack over the next 15 years.

Preparedness and Response against Biological Attacks

As a result of the terrorist attacks of September 11, 2001 and the attacks with anthrax contaminated letters, there has been increased emphasis on the need to develop capabilities to prepare for current and future bio-wars and terrorist attacks, especially potentially catastrophic attacks by terrorists using biological weapons.

US CDC developed the strategic plan for the purpose of overall planning to upgrade the national public health capability for responding to biological and chemical terrorism. This plan is based on the following five focus areas, with each area integrating training and research: 1)Preparedness and prevention. 2)Detection and surveillance (Early detection is essential for ensuring a prompt response

to a biological or chemical attack, including the provision of prophylactic medicines, chemical antidotes, or vaccines). 3)Diagnosis and characterization of biological and chemical agents. 4)Response (A comprehensive public health response to a biological or chemical terrorist event involves epidemiologic investigation, medical treatment and prophylaxis for affected persons, and the initiation of disease prevention or environmental decontamination measures). 5) Communication.

Meanwhile, at the outset of military operations, US DoD has three broad goals: 1)If possible, prevent an adversary use of chemical or biological weapons in the United States or abroad, 2) provide rapid and uninterrupted force preparation and deployment in the face of CB threat or use, 3) provide comprehensive force protection while accomplishing the mission. There are three principles in biological defense. First, no one technology or set of procedures is sufficient to counter the threat of chemical and biological weapons. Second, to provide the most effective approach to avoid contamination and sustain operational tempo on an asymmetric battlefield, the military response is based on a system of systems, which are organized according to three principles : Contamination Avoidance, Protection, Restoration (Decontamination and Medical Countermeasures). Third, these three basic principles are supported by a variety of other tools : Modeling and Simulation, Warning and Reporting, and Command and Control.

In both cases, civilian and the military, one of the important aspects of the response to biological acts when they involve deadly microorganisms is the speed with which the response plan goes into effect. Delay results in greater numbers of infected people and much more economic burdens.

The distinction between terrorist and military use of BW is increasingly problematic. The stealthy qualities of biological weapons further complicate the distinction between terrorism and war. In reality, all biological attacks are likely to require an integrated response involving both military and civilian communities. Despite this complication, however, there exist distinct differences, due to the very nature of a civilian population, between military and civilan goals to establish an effective biological countermeasures. The focus of civilian goals for response is a prepared and ready response force.

Biological weapons differ in several important respects from other weapons of mass destruction, and thus require a different approach for deterrence, detection, and response. Understanding these differences described above is critical to formulating public policy.

Research and Development of Biodefense Technolgy

This section outline the R&D program for biodefense mainly focused on the US DoD plan. Emphosis is mainly on the Contamination Avoidance (Biological Detection) and Medical Systems areas.

- Contamination Avoidance (Biological Detection)

The goal of contamination avoidance is to provide real-time capability to detect, identify, characterize, locate, and warn against all known or validated BW agent threats below threshold effects levels. Earliest possible warning is a key to avoiding biological contamination. research and

development efforts seek to optimize and balance system sensitivity, size/weight, cost, power consumption, signature and false alarm rate. Ultimately the goal is direct integration of CB detectors as a single system into various platforms, and command, control, communication, computer, and intelligence(C4I) networks. Ongoing Defense Technology Objectives (DTOs) in this area are the follow:

- Biological Sample Preparation System for Biological Identification
- Stand-off Biological Aerosol Detection
- Chemical Biological Agent Water Monitor
- Biological Warfare Defense Sensor Program
- Activity-Based Detection and Diagnostics
- Force Medical Protection/Dosimeter ACTD
- Terrorist Chemical/Biological Countermeasures

DoD's biological defense science and technology efforts in this area is divided to two categories. One is a point detection and the other stand-off detection. The applicability of BA detection equipment to emergency first responders will be dependent upon the characteristics of the detection equipment, as well as the type of biological agent to be detected and the objective of the emergency first responder unit. Because of the complexity of the environment and the need for high selectivity and sensitivity, the biological detection systems are necessarily complex devices consisting of various subunits., i.e., multi-component analysis system.

Based on a system of systems, biological agent point detection systems generally consist of four components as shown in the Biological Integrated Detection System (BIDS): the trigger/cue (nonspecific biological agent detectors; particle measurement and/or fluorescence methods), the collector, the detector: wet(flow cytometry) or dry detection (mass spectrometry), and the identifier (antibody-based identifiers and nucleic acid amplifier).

Standoff technology uses the concept of detecting and measuring atmospheric properties by laser remote sensing or LIDAR for remote early warning of BAs. IR based LIDAR systems are able to see out to ranges of 30 km to 50 km as the atmosphere is fairly transparent to this wavelength of light. One of limiting factors to standoff systems is the lack of availability of small, inexpensive high-power lasers.

Military biological detection systems are currently in the research and early development stages. There are some commercially available devices that have limited utility and are generally high cost items.

Three of the key biological detection systems fielded today are BIDS-NDI, Portal Shield, and the Biological Weapons Agent Sampling Kit.

Contamination avoidance modernization strategy in biological defense area include: Joint Portal Shield providing an automated network biological detection capability to protect high value fixed sites; Automatic long line source and point/mobile Biodetector to detect and identify BAs(JBPDS); Navy-Ship based Interim Biological Agent Detector(IBAD); and Army-Biological Integrated Detection System (BIDS).

Currently, the Joint Program Office for Biological Defense (JPO-BD) also manages the biological

detection efforts, such as Joint Biological Standoff Detection System (JBSDS), Critical Reagents Program(CRP), and Technology Transfer Program. The CRP consolidates all DoD antibody, antigen and gene probe/primer developments and requirements. The CRP is tasked with ensuring the availability of reagents critical to the development, test and operation of biological defense systems; supporting research, development and acquisition efforts to ensure the best possible reagents are available against current and emerging threat agents and producing Hand Held immunochromatographic Assays (HHAs) and DoD Biological Sampling Kits.

Current sensor and system R&D technologies are usually grouped in three catagories: Detection and identification, reagent/assay development, and supporting technologies. Detection and identification technology include the following devices: PCR for genetic detection of bacterial and viral agents, microchip platform for detection, mass spectroscopy methodologies for sample handling/analysis, immunobead force differentiation assay, pyrolysis-GC/ion mobility spectrometry, optical particle classifier, and amplifying fluorescent polymer. Supporting technologies that will eventually contribute to the bio point detection technologies are ambient background characterization, aerosol sampler development, and threat agent characterization.

There are four related programs currently ongoing within DARPA that contribute to the development of advanced sensor technology: BW Defense Environmental Sensors, Tssue-Based Biosensors, Microfluidic Molecular Systems, and Pathogen Genome Sequencing.

The emergency first responder involved in the early stages of crisis management, must recognize that while public health laboratories and supporting clinical laboratories have the capability to detect and identify possible BAs, these tests are usually not field deployable. Generally, the laboratory-based systems are slower than field systems, but exhibit greater selectivity and versatility than field-based.

BAs are effective in very low doses. Therefore, BA detection systems need to exhibit high sensitivity and selectivity, and rapid response time. The major technical challenges are in the areas of biological detection system including remote/early warning sensing, improved agent discrimination and quantification, sample processing, interferent (i.e., false positive and negative alarms) and ambient biological background rejection, and genetic probe development. Size, weight, and power reduction of detectors, power generation and consumption, development of integrated biological and chemical detection systems, and the fusion of sensor data with mapping, imagery, and other data for near real-time display of events are other areas of challenge. There are two critical needs focused on biological agent detection. Current technologies require a high level of logistical support and lack discrimination in biological standoff detection.

- Protection (Non-Medical Protection)

The focus of protection is to prevent exposure or the effects of exposure, and includes medical capabilities, such as vaccines. Protection includes all medical and non-medical means. There are two types of non-medical protection: Individual(protective masks and clothing.) and collective(various types of NBC protective filters, entry/exit, and air movement devices). Because the primary route of exposure for BAs is by inhalation, non-medical protection includes efforts to prevent exposure to or

the effects of BAs. A variety of protective masks have been developed and fielded to protect individual from exposure. To protect against exposure as a result of contact, various protective clothing items, including suits, boots, and gloves, have been fielded and advanced systems are under development. Within the science and technology base, the ongoing DTOs are Advanced Adsorbents for Protection Applications and Self-Detoxifying Materials for CB Protective Clothing which detail key protection efforts.

Major technical challenge in non-medical protection program is that integrating CB protection into future weapon systems necessitates tradeoffs between performance requirements and limitations of materials and designs.

- Restoration (Decontamination)

Restoration capabilities include medical and non-medical measures required to restore the joint force, units, facilities, and equipment to near-normal operating conditions after being challenged by a biological agent hazard. These measures include non-hazardous decontamination operations, effective supply and sustainment of all defense assets, and effective medical diagnostics and post-exposure countermeasures required to allow rapid determination of agent exposures and subsequent treatment.

In this section, only the decontamination (disinfection in bioscience) area is outlined and medical measures are referred in the next medical systems section.

Decontamination is organized into three categories that reflect operational urgency: immediate, operational, and thorough decontamination, and also entails special considerations for patients, sensitive equipment, aircraft, fixed sites, and the retrograde of equipment. In US DoD, there are three key development efforts: Joint Service Sensitive Equipment Decontamination, Joint Service Fixed Site Decontamination System, and the Superior Decontaminant System. Decontamination R&D programs are technologically divided into three major areas including Solution Phase Chemistry, Gas Phase Chemistry and Supporting Technologies.

Decontamination efforts draw on an extensive array of basic research and supporting technologies. Current decontaminants cause adverse effects to physical, optical, electronic, or mechanical properties of the items being decontaminated and are not environmentally friendly. Some of the technologies being explored to address these limitations include material survivability technology, supercritical fluidics, decontaminant coating technologies, thermal desorption methodologies, gas phase decontamination, chemical matrix strategies, and novel approaches using non-ozone depleting solvents, plasma, oxidation catalysts, peroxycarboxylic acid, novel surfactants and microemulsions, dioxiranes, and nanoparticles. To reduce dependence on water, non-aqueous technologies are being explored, including gas phase decontaminants, destructive adsorption, and organic chemical matrix strategies. A critical challenge is personnel and patient decontamination. Enzymatic decontamination, antimicrobial nanoemulsions, skin and wound decontaminants, and other methods for personal decontamination that does not harm the individual are being explored.

An information guide, "Guide for the Selection of Chemical and Biological Decontamination Equipment for Emergency First Responders, NIJ Guide 103_00 Volume I, Oct. 2001," developed by

the National Institute of Justice through literature searches and market surveys, provide emergency first responders with information that should aid them in the selection and utilization of chemical and/or biological decontamination equipment.

There are two principal technical difficulties associated with this effort. The first is the development of decontaminants that are reactive, non-aqueous, non-corrosive, safe for use on sensitive equipment, able to decontaminate a broad spectrum of chemical and biological agents for dual-use, environmentally safe, and pose no unacceptable health hazards. The second technical difficulty is the development of decontamination systems that effectively clean all surfaces and materials, while at the same time reduce the manpower and logistics burden.

- Medical Systems

Along with individual and collective protection, medical systems forms a third area associated with the NBC defense principle of protection. Medical Systems include all pharmaceuticals, biologics, and devices that preserve combat effectiveness by timely identification, diagnosis, and provision of medical countermeasures. These products are critical elements of medical biological defense, and provide the ability to protect the human-beings from BAs, to rapidly diagnose (in clinical specimens) infection from agents, and to treat casualties.

Under the CBDP, the Joint Medical Biological Defense Research Program (JMBDRP) is chartered as the joint focal point for medical research efforts to counter BW threats, and thus responds to requirements from the DoD. The JMBDRP includes the following areas of development: Pre-exposure countermeasures and post-exposure countermeasures against bacteria, viruses, and toxins. Also being developed are rapid portable diagnostics that will facilitate a quick medical response for exposed warfighters. Currently, the most effective countermeasure is pre-deployment active immunization.

Currently licensed and IND solutions for use in medical biological defense include vaccines and antisera against: Anthrax, botulinum toxin, small pox, Q fever, tularemia, and three equine encephalitis viruses. In mid and far-term, according the Biological Defense Modernization Strategy, emerging medical biological defense products will be developed as follows: Recombinant plague vaccine, next generation anthrax vaccine, multivalent Venezuelan equine encephalitis vaccine, recombinant staphylococcal enterotoxin multivalent vaccine, recombinant ricin vaccine, antibiotics and antiviral Drugs, and Joint Biological Agent Identification Diagnostic System (JBAIDS; PCR-based and immunodiagnostic systems).

Highly sophisticated technology base efforts hold the promise of yielding important new products and technologies against a wide range of biological threat agents. These products include multi-agent vaccine and its delivery capabilities/systems that will reduce costs of vaccine production and simplify immunization schedules, and a common diagnostic system that can be deployed at forward sites to rapidly analyze clinical samples for the indications of BW agents as well as infectious diseases of military importance. The current JMBDRP includes the following research areas for the development of medical countermeasures:

-Characterize the biochemistry, molecular biology, physiology, and morphology of BW threat agents.

-Investigate the pathogenesis and immunology of the disease.

-Determine the mechanism of action of the threat agent in animal model systems.

-Select antigen(s) for candidate vaccines.

-Develop and compare potential vaccine candidates and characterize their effects in animal models.

-Develop surrogate markers of efficacy.

-Establish safety and efficacy data for candidate vaccines.

-Develop medical diagnostics to include far forward, confirmatory, and reference labs.

-Develop chemo/immunotherapeutic agents and preparations

The complementary technology-based research and development to enhance and expand these capabilities and to identify and develop new capabilities, is also being supported by collaboration with other agencies, including the Defense Advanced Research Projects Agency(DARPA) and the Department of Energy. Selected DARPA research efforts in the Unconventional Pathogen Countermeasures Program and Advanced Medical Diagnostics Programs are as follows:

-The development of broad-spectrum vaccines by molecular breeding (gene shuffling) strategies.

-Broad-spectrum antimicrobial drug discovery research.

-High-level plant-based expression system for vaccine antigens and epithelial transport molecules.

-The development of a proprietary B-cell sensing technology for rapid and sensitive medical.
diagnostics for biological threat agents and endemic diseases.

-Development of in vivo countermeasures against biological toxin threats of the superantigen family,
exemplified by staphylococcal enterotoxin B (SEB), using a peptide or peptidomimetic antagonist.

In addition to these technologies, a variety of other technologies are being also developed. Key technologies among them include recombinant vaccine development efforts (e.g., gene insert, gene shuffling techniques), immunomodulators to provide enhanced immunity against any pathogen, active and passive immunoprophylaxes, novel genomic, molecular genetics, molecular phylogeny, active site-directed inhibitors, receptor antagonists, and small molecule antibiotics and protein inhibitors.

The use of animal models remains a critical aspect in the development of some medical products. One of the challenges in the development of some medical products is a continuing and growing lack of availability of specific non-human primates. DoD is currently investigating the total non-human primate requiements, and identifying alternative models, including non-human primates other than those in short supply, other animal models, and non-animal models (e.g., cell cultures). This investigation is intended to preclude potential resource limitations from slowing the development of medical NBC defense products. In the private sectors, in addition to the technical challenges in DoD, the lack of high-level biological containment (BL-3 and BL-4) laboratory facilities, and the lack of widespread scientific expertise in biological defense, are the critical barriers.

Conclusion

The deliberate use of microorganisms and toxins as weapons has been attempted throughout history. Thus, the threats posed by biological weapons are likely to continue into the future.

Biotechnology is moving fast. All the developments in biotechnology will give more and more information about how to develop and manufacture sophisticated types of biological weapons. As a result, an enemy's ability to produce genetically engineered threats on demand exacerbates the long-lead time between research for a medical solution and obtaining FDA licensure for the medical product. However, preparing the nation to address this threat is a formidable challenge.

Accelerating effforts to improve disease surveillance, and research and development of improved and emerging diagnostics, therapeutic agents and vaccines will promise to serve the dual purpose of protecting the public health and defending against biological weapons. By creating and supporting an infrastructure comprised of training and education, surveillance, early warning, and communication networks, the frontline responders will be better prepared to recognize and respond to acts of bioterrorism. To cope with the technical barriers and to enhance our biodefense capabilities, large-scale international cooperation should be accompanied.

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