Biological warfare: The many possibilities

Wasserman E
Department of Medical Microbiology, University of Stellenbosch

Correspondence: ew@sun.ac.za

Biological warfare has been a threat to society for many decades. This article summarises some of the most likely organisms suitable for a bio-terrorist attack, including Bacillus anthracis, Francisella tularensis, Yersinia pestis, the agents of viral haemorrhagic fevers, Variola virus and botulism toxin. ***(SA Fam Pract 2004;46(8): 40-42)***

Summary
The threat of biowarfare has been highlighted following the 11 September 2001 attack on the World Trade Centre. Although disease as a weapon of destruction is not a modern concept (for example: medieval warriors catapulted corpses of plague victims over the walls of beleaguered cities), modern microbiological methods provide the means for the mass production of lethal infectious substances in a form suitable for wide dissemination. Molecular engineering can enhance the ability of infectious agents to endure in the environment, be more virulent and express increased resistance to antimicrobial agents. Some of the more likely agents suitable for use in biowarfare are briefly discussed.

Requirements for suitable biological weapons

Bioterrorism is defined as the use of biological agents to inflict disease and/death on humans, animals or plants.

The following requirements should be met in order for an infectious agent to be an effective biological weapon:

- The agent must be producible in sufficient quantity and in a stable form.
- It must be possible to deliver the agent in a way that would affect the maximum number of targets via the most effective route.

Possible routes of dissemination of infective agents include:

- Contamination of food or water supplies
- Dispersion in aerosols (air-borne route). This route is suitable for infective agents as well as toxins. Particle sizes of 1-10 μm are desirable. Under ideal conditions, these particles may remain suspended in the air for hours and are sufficiently small to make their way into the distal bronchioles and terminal alveoli after inhalation. Low-cost, easily obtainable equipment can be used for production.
- Percutaneous administration – this route is less practical to target a sizeable population.

Biotechnological methods for genetic engineering of biological weapons can be employed to augment their potency:

- Expression of toxins
- Increased virulence
- Acquisition of resistance to antibiotics
- Prolonged survival rates during storage and aerosolization.
- Combining organisms (e.g. hybrid between smallpox and Ebola viruses was allegedly created by Russian researchers).

Candidate agents for bioterrorism

Bacillus anthracis (anthrax)

This is a zoonoses occurring widely in nature, and can infect all mammals. Following the death of an infected animal, spores are formed which are extremely hardy and may persist in the environment for up to 40 years. Infection in humans follows exposure to infected carcasses or other infected animal products.

Bacillus anthracis forms large Gram-positive rods, with oval subterminal spores. Spores are formed only in the presence of oxygen, and are not always visualised in clinical material. The organism is easy to isolate and grow in the laboratory.

Clinical manifestations:

Three forms of infection with *Bacillus anthracis* occur:

1. Cutaneous anthrax: this is acquired when a skin abrasion is exposed to infected material, introducing spores into the skin. Germination occurs within hours with the formation of toxin-producing vegative cells. A red macule develops at the site of inoculation. Characteristically, this lesion is painless. The lesion then progresses to form vesicles, followed by ulceration with a black necrotic eschar surrounded by oedema. A regional lymphadenitis frequently develops. A live-threatening bacteraemia may develop, but spontaneous healing occurs in 80-90% of untreated cases.
2. Inhalation anthrax (wool-sorter's disease): this follows inhalation of the spores, which is also the most likely type of exposure following a bioterrorist event. About 20 000 organisms can produce lethal infection if the particle size of the aerosols is less than 5 μm. The spores are phagocytised by alveolar macrophages and germinate in the alveolar spaces or in the mediastinal lymph nodes. This results in a haemorrhagic mediastinitis, massive bacteraemia and pneumonia. Meningitis may occur as a complication of bacteraemia. Inhalation anthrax may present with signs and symptoms of an upper respiratory tract infection or pneumo-

nia preceded by symptoms of severe viral respiratory disease during the first 1 to 3 days. In the acute phase, symptoms include fever, dyspnoea, stridor and hypoxia accompanied by hypotension. The chest X-ray indicates a widened mediastinum. Mortality of advanced disease is extremely high (around 90% is quoted in most reports).

3. Gastrointestinal anthrax can be contracted via the ingestion of contaminated meat that is not thoroughly cooked. Oropharyngeal or intestinal infection follows. This form of the disease is extremely rare and encountered in less than 1% of all clinical cases.

Penicillin is the drug of choice for the treatment of wild-type anthrax. Concerns about the creation of genetically engineered resistant *B. anthracis* make the use of other antibiotics such as a fluoroquinolone or doxycycline seem prudent. Although a live, attenuated vaccine is available for animal use, this is not recommended in humans. A sterile protein-based human anthrax vaccine was licensed in the USA in 1970 which will be 88% effective following a vaccination regimen comprising six subcutaneous injections over an 18-month period.

Francisella tularensis

Tularaemia is also a zoonoses, occurring primarily in the northern hemisphere. The organism naturally infects rodents. Humans may become infected after direct animal contact or via insect vectors such as ticks, biting flies and mosquitoes. The clinical manifestations of tularaemia relate to the route of exposure to the organism (cutaneous inoculation, inhalation or ingestion).

The organism may be cultured from relevant clinical material or diagnosis can be based on serology results. The aminoglycosides streptomycin and gentamicin are the drugs of choice for the treatment of tularaemia. Tetracycline and chloramphenicol may also be used, but are associated with relapse of the disease. No effective vaccine against this agent is currently available.

Yersinia pestis

Y. pestis causes plague, a zoonotic disease of rodents and other animals that is usually transmitted to humans via fleabites. This route of infection results in the bubonic form of plague, characterised by fever, malaise and a painful lymphadenitis, or a so called bubo. Septicaemia may follow. Pneumonic plague results after haematogenous spread of plague bacilli to the lungs. This form of the disease is highly contagious and can be spread from person to person via air-borne droplets. The respiratory route is also the most likely route of infection as a result of a bioterrorist event.

Early administration of streptomycin or tetracycline can reduce mortality from approximately 50% in untreated plague cases to about 5%.

Variola virus

Humans are the only natural hosts of the smallpox virus. This agent has a 3 000-year history, and is the cause of untold millions of deaths. This disease has been officially eradicated, with the last reported case occurring in Somalia in 1977. Routine vaccination against smallpox has been discontinued since 1972.

The virus may be discounted as a rational biological weapon, as person-to-person spread will be uncontrolled. Although the WHO advised the destruction of all stocks of this virus by June 1999, this was postponed until 2002 in order to allow for more research on the virus.

The usual route of infection is through inhalation of droplets containing infectious virus particles. A transient viraemia follows, spreading the virus to the internal organs and the skin. A second viraemia marks the end of the incubation period, which may last for 12-14 days, and the beginning of the toxaemic phase. Skin eruption follows within 3 to 4 days. The rash starts on the tongue and the roof of the mouth and spreads to the face, forearms and hands. Subsequently it spreads over the arms, legs, and trunk. The lesions are all at the same stage of development as they progress from macules to vesicles to pustules, and finally crusting. The disease may be controlled by vaccination.

Agents of viral haemorrhagic fevers

Viruses capable of causing haemorrhagic fever syndrome include RNA virusus of the Filoviridae (Ebola and Marburg viruses), Arenaviridae (Lassa fever, Argentine or Junin, Bolivian or Marchupo, Venezuelan or Guanarito, and Brazilian or Sabia haemorrhagic fever viruses), Bunyaviridae (hantavirus, Rift Valley fever and Congo-Crimean hemorrhagic fever viruses), the Flaviviridae (yellow fever and dengue fever viruses). Humans are exposed to these agents by contact with infected animals, or via arthropod vectors. These infections are characterised by vascular damage and altered vascular permeability. Symptoms include fever, myalgia, haemorrhages in mucous membranes, prostration and shock. A high mortality rate is associated with these infections.

Botulism toxin

Botulinum toxins (secreted by *Clostridium botulinum* strains and a few other *Clostridium* species), spread through the bloodstream and exert their effects at neuromuscular junctions, inhibiting the release of acetylcholine. The classical presentation of botulism is acute flaccid paralysis that descends symmetrically downwards from the head. Clinical syndromes consist of food-borne botulism, wound botulism and botulism caused by toxin production after clostridial colonisation of the intestines. While the symptoms, diagnosis and treatment of food-borne botulism have been well described, less is known about intoxication resulting from inhalation of toxin. The administration of botulinum antitoxin is an effective treatment, especially if the antitoxin is given before clinical symptoms become apparent.

Brucella species were recently removed from the list of most likely agents for biowarfare, but remain a possible agent along with *Vibrio cholerae*, *Burkholderia pseudomallei*, *Coxiella burnetti*, the agents of viral encephalitis and staphylococcal toxins.

References

1. Dixon TC, Meselsom M, Guillemin J and Hanna PC. Anthrax. *The New England Journal of Medicine* 1999; 341 (11): 815-825.
2. Klietmann WF and Ruoff KL. Bioterrorism: Implications for the Clinical Microbiologist. *Clinical Microbiology Reviews.* 2001; 14 (2): 364-381.

Note:

Contact INFOMED at the Tygerberg Campus Library at infomed@sun.ac.za to request one of the above references