"anonymous" in that it is intrinsically associated with, and identifiable to, only one individual (Gostin 1995).

The solution to the problem of balancing the needs for patient privacy and accessible health data is hardly straightforward. Various suggestions have been made, including the possibility of separating the contents of medical records into confidential and non-confidential portions in order to allow researchers and other third parties (such as insurance companies) access to the valuable information within (Dodek 1997, Black 1994, Seigler 1982). Other ideas include promoting a renewed prudence and awareness of the many methods by which patient confidentiality can be breached, with particular attention directed towards the hazards posed by modern technology. The growing computerization of medical records, while a convenient feature for clinicians and researchers alike, has produced yet another domain in which the confidentiality of medical records can be compromised (English 1994, Beauchamp 1991).

Although Siegler (1982) famously asserts that confidentiality in medicine has become "a decrepit concept," such a pessimistic opinion need not be the last word on this subject. Without question, physicians, researchers and patients alike must address the changing reality of patient privacy. As a result, due to the ever-increasing demand for updated, accurate health research, patients should be aware of the fact that their personal information may be shared with others for the good intentions of research. What this requires of physicians, however, is the responsibility to discuss frankly this reality with their patients, in order to ensure that they are aware of the limits of patient confidentiality. Although the reality of accessibility to medical records may be unsettling for some, ultimately patients must understand what the operating concept of patient privacy does and does not entail in the clinical and research realms of medicine. ■

Anthrax and the Three Amigos

by Ajit Thakur

Bioterrorism involves the deliberate use of microorganisms to cause infection and long-term destruction and destabilization of the human population.

The notion of Bioterrororism was conceived during World War I and II by the Germans, Japanese, Americans, Russians and the English. Only recently has Iraq developed the powder form of this microorganism. Even Canada had once actively participated in the development of weapons grade Anthrax spores for Britain. At the end of WWII, hundreds of pounds of Anthrax spores were barrelled and cast away into the depths of the St. Lawrence River in Canada.

There are thirty different pathogenic microbes used in Biological warfare. They include viruses, bacteria, toxins, and animal venom eg: Anthrax, *Clostridium botulinum*, Plague, Smallpox virus, and Tularaemia.

The anthrax bacillus, *Bacillus anthracis*, was the first bacterium shown to be the cause of a disease. In 1877, Robert Koch incubated pure cultures of the organism and demonstrated its ability to form endospores. He was the first to experimentally induce anthrax by injecting it into animals. It was later found that *Bacillus anthracis* is a gram-positive bacterium. It is non-motile and multiplies as vegetative rods capable of spore formation that enable it to resist extreme environmental conditions.

The Anthrax bacteria and the spores are present in the soil in all developing and developed countries. It is endemic, most commonly present in the soil of Asia, Middle East, Africa and even some places in the United States, like Texas, Oklahoma, Minnesota, Dakota and Nevada.

Grazing animals commonly acquire this disease. When the animal ingests the bacterium, the poison of the anthrax kills the animal cells. Once the animal is dead, the spores and the vegetative rods multiply and spread from the carcass into the soil and then are spread by the wind and soil into the environment. This is the only way through which the bacterium propagates itself.

It might be surprising to note that Anthrax bacteria are actually harmless themselves, if not for a few mutations that sets them apart from the others. An individual bacterium can be

compared to a terrorist state, which harbours three infamous proteins whose teamwork is unlike any seen before. Together, these three amigos are capable of manipulating cellular machinery into reprogrammed self-destructing units.

These three amigos are aptly named:

1. Protective Antigen (PA)
2. Edema Factor (EF)
3. Lethal Factor (LF)

Site of infection	Pathology
Cutaneous Anthrax infection	The bacteria first enter an open wound on the body to attack a foothold in the new tissue, and it is here that the immune system is most likely to be recognized early.
Gastro-intestinal infection	When the spores in the human host reached the bacteria in the spores have colonized and they enter the body and damage the gastrointestinal tract, causing bleeding and death.
Inhalation Anthrax (woolsorters' disease) Highest fatality rate/dose	When anthrax spores are inhaled in the breathing of the spores, they travel to the mediastinal lymph glands where the bacteria cause bleeding in the lymph nodes, leading to death.

Inhalation Anthrax is the most common bioterrorist weapon. Symptoms usually develop in seven days as flu like feelings including lethargy, runny nose, headache, fever, cough and sweating. Two days after the symptoms, the patient develops respiratory failure and the mediastinal chest lymph glands swell, which leads to bleeding inside the lungs, soon followed by patient death.

Japan used anthrax spores against Chinese in Manchuria during World War II in the 1940s. Germans used it against Jews and their enemies in World War I and II. Aerosol dispersed anthrax spores were once the preferred biowarfare agent used in controlling enemy soldier populations. Recently, the World Health Organization reported that a powdered aerosol anthrax spore attack even today would unquestionably be deadly to the masses.

Current Research

When *B.anthracis* spores are inhaled, they become ingrained into the lungs, where the body's immune system sends security guards called macrophages to ingest them. However, these unsuspecting cellular security guards (macrophages) are being used as a spore-transport vehicle to attain access to the secure distribution centre (lymphatic system) of the body. Unlike other ingested organisms, these spores are not destroyed, and are free to germinate and multiply within membranous compartments called phagolysosomes in macrophages. The spores then form vegetative cells that alter cellular processes leading to cell death. However, this process of vegetative propagation has been carefully orchestrated and ensures that sufficient time has elapsed for the macrophage to migrate to the lymph nodes, where the mature bacteria are released. At this point the bacteria have a direct route into the bloodstream, where they are equipped to proliferate unhindered and begin to secrete their infamous anthrax toxin.

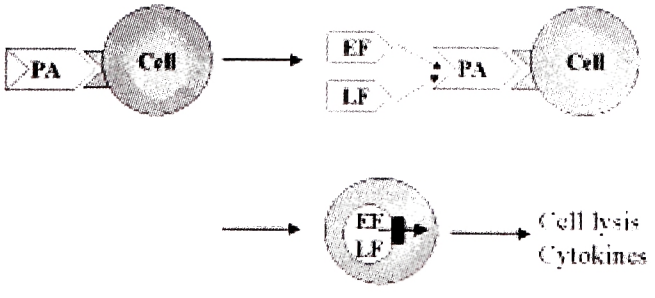

Figure 1:

Bacillus anthracis toxin at work. Edema factor and Lethal factor use the Protective antigen to gain access to the insides of a cell. Its subsequent actions lead to Cytokinesis.

It turns out that *B. anthracis* is itself no different from other harmless, soil-dwelling bacteria were it not for the two extra DNA molecules known as pXO1 and pXO2. pXO2 codes for the proteins that form a protective capsule around *B. anthracis*. On the other hand, pXO1 encodes for several proteins including the deadly trinary arsenal of toxin it formulates. The toxin turns out to consist of three proteins: Protective antigen, edema factor and lethal factor. These proteins are the three amigos that act cooperatively to slip past cellular defences. For example, the edema factor present in the bloodstream immediately disables a key protein involved in calcium-triggered signalling pathways, and uses it to stimulate its own catalytic activity. The resulting activity inhibits the body's immune response against the bacterium, thereby permitting their proliferation. Despite the bacterium's progress, researchers have implied that the bacterium is essentially harmless until this stage of the infection, where they have not yet begun to attach and penetrate cells. It is now that these amigos can begin to sabotage cellular machinery into submission.

First, the protective antigen binds to the surface of a cell, where it is activated by an enzyme (protease) that cleaves its tip. Seven of these cleaved molecules aggregate to form a ring shaped structure known as a heptamer, that subsequently binds to varying numbers of edema and lethal factors. The receptor for the protective antigen then aids this complex, consisting of the heptamer bound to the two factors, in their transport into an internal membrane-bound compartment called an endosome. Mild acidity causes the heptamer to embed itself into the compartment's membrane where subsequent conformational changes leads to the transport of the edema factor and lethal factor across the endosomal membrane into the internal matrix of cells. After a successful entry, the two factors proceed to hijack the cell. In essence, the heptamer is like a door, between the endosome and the cellular matrix, which opens to provide a passage for edema factor and lethal factor in response to the slight acidity of the endosomal environment.

Once inside the cell, edema factor and lethal factor catalyze different molecular reactions. Edema factor upsets the controls on ion and water flow across cell membranes and thereby promotes the swelling of tissues. In phagocytes it also saps energy that would otherwise be used to engulf bacteria.

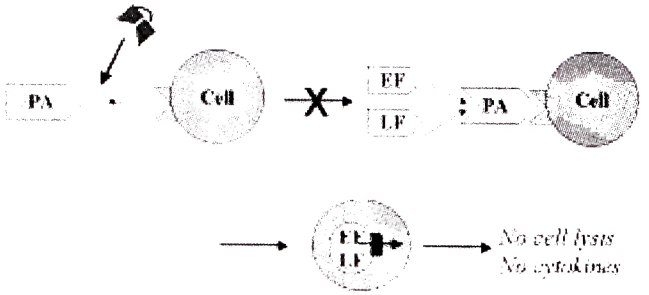

Figure 2:

Neutralizing Antibody impeding the action of Bacillus anthracis toxin. This successful approach relies on using neutralizing Antibodies to bind to the Protective Antigen and inhibit its action. Thus, Edema factor and Lethal factor will not gain access the cellular interior to cause cell lysis.

Above from: www.che.utexas.edu/.../Research/ researchpic/anthrpic.htm

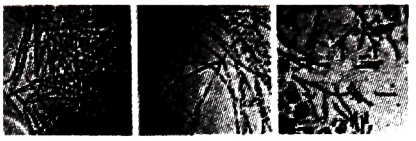

Figure 3: Robert Koch's original micrographs of the anthrax bacillus

Anthrax is primarily a disease of domesticated and wild animals, particularly herbivorous animals, such as cattle, sheep, horses, mules, and goats. Humans become infected incidentally when brought into contact with diseased animals, which includes their flesh, bones, hides, hair and excrement.

Above from: www.bact.wisc.edu/Bact330/ lectureanthrax

The focus of current research is to find a cure and effective control for this potent disease. There are three viable means of preventing an epidemic of anthrax. One approach uses antibiotics to kill the bacteria, the source of the problem, which produces the toxin. Some common antibiotics are: Penicillin and Ciproflaxin. However this approach fails to take into consideration any toxin that has already been produced, leaving the toxin to wreak havoc in the body. This approach will only be effective if implemented early. In conjunction with anti-toxins, this form of treatment constitutes the second approach to battling an anthrax epidemic. Another pre-emptive approach involves use of a vaccine to neutralize the effect of the anthrax toxin. For example, Anthrax Vaccine Adsorbed (A.V.A.) is currently being administered to American soldiers as a preventative shield to combat anthrax. This anthrax vaccine works by stimulating the human immune system to produce antibodies that bind to the toxin and cause protective immunity. Livestocks in parts of US are routinely given anthrax vaccine. However, due to limited supplies, this approach cannot be applied on a large scale, when it becomes necessary if the entire population is to be protected against the toxin.

Waging War at the Cellular level

There are three primary treatment ideas. One innovative approach involves the use of decoy receptors that prevents the protective antigen from linking to its receptor on the cell. These decoys can be introduced as soluble copies of the toxin receptors' protective antigen-binding site called sATR. The next approach involves distancing edema factor and lethal factor and preventing their attachment to the binding sites on protective antigen heptamers. This can be achieved by plugging heptamer sites, the entrance doors to the cellular interior, with linked copies of a molecule that also has affinity for that site. This idea is effective, however, these cellular terrorists are free to find any of the innumerable number of other unprotected and unplugged heptamers to bear the brunt of the attack. The last approach also hinges on the idea that involves blocking the transport of edema and lethal factor from the endosome into the cytosol. This can be achieved by incorporating a version of protective antigen known as a dominant negative inhibitor, DNI, into newly formed heptamers thus inhibiting them from moving edema factor and lethal factor from moving across the endosome's membrane. In contrast to the previously stated method, this approach will effectively catch these amigos in the act of sabotage, thus preventing any further terrorism on other cellular nations. Such attempts to foil the toxins' ability to function effectively are the key to developing an anthrax vaccine and are currently being researched at the leading research institutions of North America.

Figure 4:

This picture shows the effects of the three main types of Anthrax: Cutaneous, Gastrointestinal and Inhalation.

Above From: http://www.anthrax.osd.mil/disease/infection.asp

The latest research has aimed at producing a large-scale effective vaccine and an antidote for the anthrax toxin. Some of the most successful methods have focused on producing mutant toxins as cellular competition for the real toxins. However, the mutants lack the operational ability of the real toxin. Thus, these mutant toxins plug the heptamers, making them unavailable for the real toxin. Other methods aim to neutralize the anthrax toxin using antidotes.

In any case, bioterrorism has opened a new chapter in science and medicine. To combat the threat of bioterrorism is to be aware of the multiple agents that can be used to counter-react and control against such acts. It would be best for a society to be prepared to combat and manage the overall impact of such serious destruction. There is no doubt that the current research on the biology of *Bacillus anthracis* and on possible therapies and vaccines will one day provide a range of effective therapies for anthrax treatments. It is fervently hoped that every human life will be spared from infection and death due to biological terrorism, which in fact is the very essence of medical research. ■

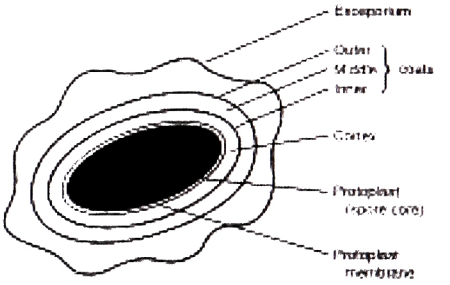

Figure 5:

The multiple layers of protection shield the anthrax bacterium from extreme environmental conditions and maintain its dehydrated dormant state. .

Picture above from: http://www.gsbs.utmb.edu/microbook/ch015.htm

Figure 6:

Bacillus anthracis produces the deadly anthrax toxin.

Found at http://www.nature.com/nsu/011004/011004-9.html