Developing vaccines to counter bioterrorist threats

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Large and innovative research programs are underway to define the immune parameters for vaccines against a wide array of pathogens considered to represent a potential bioterrorist threat. However, the development and utilization of such vaccines presents a number of predicaments that have not previously been addressed by the field of vaccinology.

The beginning of the 21st century sees the field of vaccinology in a state of confusion that would make Jenner turn in his grave. In the developing world, there remains a desperate need to broaden availability of tried-and-tested vaccines against common, childhood pathogens in programs that could save millions, if only there was the will, organization and funding to carry them out. In many parts of the developed world, diseases such as polio, measles and mumps, once thought to be largely controlled as a consequence of public health vaccination programs, are re-emerging in the face of public complacency about herd immunity and suspicion over side effects. The need to develop vaccine programs against potential bioterrorist and biowarfare pathogens, however, poses a uniquely challenging set of questions: which agents we need protection from; who the target population to vaccinate is; what form effective vaccines would take; and how we will persuade people to take vaccines against the unlikely eventuality of attack, when many are suspicious of vaccines, even against sure-fire endemic infection. Furthermore, if we are aiming to vaccinate against a potential outbreak of unknown and unknowable probability, this raises the dilemma of how to properly conduct an ethical assessment of the balance between side effects of the vaccines themselves and the benefits to be gained from them.

A number of problems, of course, relate to the nature of the pathogens under investigation and, often, the lack of good animal models in which one can faithfully analyze responses to them. A major problem is how to model the disease process sufficiently using in vivo models to allow a reasonably accurate estimation of likely efficacy of the candidate vaccine. Since the vaccine targets are potent human pathogens, no test of efficacy of the candidate vaccine in humans is feasible and, therefore, there is a heavy dependence on the identification of immune correlates of protection as surrogate markers of efficacy. This sets these agents apart from those of traditional public health concern that cause regular or sporadic outbreaks in the population. In most cases, researchers lack a clear notion of the immune correlates of protection that should be sought in clinical trials. For various agents, the parameters may encompass defined CD4 or CD8 epitope T-cell responses; responses of particular profile with respect to cytokine release; and/or neutralizing antibodies of particular specificities.

The deliberate release of anthrax spores via the US postal service in 2001 emphasized a number of key points regarding the challenges posed by bioterrorism. First, even a relatively small-scale attack has the ability to create massive and disproportionate international alarm among the public, making biologic pathogens potent tools for terror. Second, the position of a democratic government with no coherent contingency plan for dealing properly and specifically with any such attack would be untenable. Third, by its very nature, terrorism is unpredictable in the individuals targeted;
many hundreds of thousands of military personnel had been immunized against anthrax, yet the individuals affected in this case were all unimmunized civilians. Lastly, while the actual number of victims was relatively small, the amount of suspicious material that had to be screened and the number of people indirectly affected by the events was enormous [1].

The US and other governments are preparing very robust countermeasures against bioterrorist agents, not least through the dedicated funding programs undertaken through the National Institute of Allergy and Infectious Disease (NIAID). In the next few years, a great deal of immunologic research will be carried out to advance the prospects for potent vaccines. The aim of this article is to analyze some of the complex issues that are under discussion in bringing these countermeasures to bear on the bioterrorism problem in an effective way.

Which pathogens?

The terror of the bioterrorist, as with any other terrorist, lies in the unpredictability of their modus operandi. Should the list of pathogens with which we are most concerned be limited to those that we know have previously been weaponized for use in a biowarfare setting, such as anthrax and tularemia? Should the list extend to other agents that are most vivid in the public consciousness as potentially lethal agents, such as smallpox and plague? Should special attention be paid to agents such as staphylococcal enterotoxin B (SEB) and other bacterial toxins, which can be synthesized without the need to have access to traceable, biologic stocks? In fact, the list of US Center for Disease Control (CDC) category A–C pathogens takes an inclusive view, encompassing the widest possible range of agents that could cause widespread death or incapacity (BOXES 1, 2 & 3) [101].

The list ranges from filoviruses, such as Ebola and Marburg, to bacteria such as Shigella. Thus, an enormous additional dividend from the biodefence research programs is the detailed analysis and development of therapeutics for a wide range of natural infections, many of which cause considerable mortality and morbidity in countries of the developing world [2]. Indeed, in a broader context, the biodefence programs can be viewed as bringing about an accelerated coming of age for molecular immunology. There has been a need for rapid progression from a preoccupation with the analysis of epitopes in a small number of model antigens, to facing up to the application of this basic knowledge for the analysis of the full pathogenic menagerie that poses a challenge to humankind.

Since the Second World War and then the Cold War, anthrax has loomed large in the public consciousness as an agent of biowarfare, owing to the perception of a rapid and painful death in the absence of early access to antibiotics, and the notion of pathogenic spores which can be spread easily as airborne particles and then linger in the ground for decades. In 1942, feeling the need to prepare for the potential use of biologic agents by Nazi forces, the British Army released anthrax spores onto the remote Gruinard Island off the coast of the Scottish Highlands, rapidly killing the island’s resident sheep. It was not until some 50 years later that a contract was issued for heavy-duty decontamination measures on the island, and a fresh herd of sheep was introduced to demonstrate that the process had been successful. The penetrance and potency of anthrax spores was re-emphasized more recently in the attack through the US postal system, in which one of the victims had received a letter which had become minimally contaminated simply by passing through the same sorting machine as one of the deliberately contaminated envelopes. The events of that time also emphasize the enormous challenge to our monitoring systems of being able to detect an attack, characterize it, and then, as alarm mounts, put into place large-scale screening that can accurately identify genuine cases without scoring false-positives or -negatives [1].

Several recent papers have modeled the potential impact of an anthrax attack in the face of different levels of response [4–6]. The analysis by Brookmeyer and colleagues concludes, “The quantification leads us to a realization that the destructive capability of weaponized anthrax is equivalent to that of a nuclear bomb... The model assumes a point-release of 1 kg of spores, concentrated at a trillion spores per gram, from a height of 100 m, in a city of 10 million inhabitants...” The conclusions drawn from computer simulations of the model are stunning, even in the base case ($p = 0.0$), when the post-attack response is relatively efficient. In the base case, more than 100,000 deaths result in the population of 10 million inhabitants. Less aggressive distribution of antibiotics to asymptomatics ($p > 0.0$) increases this number up to sevenfold” [4].

Detailed evaluation of the relative risks posed by different agents is necessarily a combination of military intelligence, rumor and precedent. The history of bioterrorism and evidence for the existence of stockpiles of a range of pathogenic agents has recently been excellently reviewed elsewhere [7]. Certainly, anthrax and smallpox feature strongly in these appraisals because there exists detailed information regarding their past.
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Who to immunize & when?

It will be clear from the preceding discussion that, in establishing a program of countermeasures to defend against bioterrorist agents, there are difficult questions to be faced regarding which vaccines we need, who should receive them and when, and how one goes about a proper cost-benefit analysis for such a program. Through the developing history of vaccinology, the conceptual framework for each of these issues has tended to be less challenging than the one we currently face. Where there have been common infectious agents that pose a significant threat in terms of morbidity or mortality, and a safe, tested, prophylactic vaccine is available such that the risks from the infection considerably outweigh any risk from side effects of the vaccine, arguments for the benefit of vaccination have been clear. Vaccination against bioterrorist agents, however, carries this debate into new areas. The first question to consider is when to immunize. These questions have often been considered with respect to the issue of the threat of smallpox. This case involves a highly infectious and lethal pathogen for which the cost of inadequate countermeasures, particularly with respect to immunity of healthcare professionals, would be very high. There exists a tried-and-tested vaccine that can, and has, been stockpiled in relatively large amounts. However, there are clearly documented side effects of this vaccine such that, were one to reintroduce very widespread public immunization in the absence of any clear and specific threat of smallpox attack, the number of individuals seriously harmed by the vaccine would outnumber those seen to be saved from a terrorist attack, and public confidence in the countermeasures would be severely shaken [8]. The first compromise is to limit the number to be vaccinated in the first instance, focusing on first-line responders such as medical and military personnel. Clearly though, this sidesteps to some extent the issue of an effective countermeasure to the release of smallpox in a subway system, for example. The next compromise is to argue that mass immunization would be part of the immediate response to any attack. Certainly for an agent such as smallpox, such a measure would be vital to avert fears of an epidemic spreading through the population. For many of the very rapidly acting agents on the category A–C pathogens list, this strategy may have a sense of closing the stable door after the horse has bolted. In the case of anthrax, which would kill within days in the absence of immediate antibiotic cover and yet is not highly contagious, it is harder to argue the effectiveness of an immunization program that might take several boosts over several weeks or months to elicit protective antibodies, unless the fear was of a protracted, ongoing bioterrorist attack. Nevertheless, a case can be made in favor of some effective benefit accruing from postexposure immunization programs in combination, where necessary or appropriate, with antibiotics [9].

Through recent conflicts, the biodefence appraisal has been that military personnel may come under attack from biologic weapons, so that these individuals may be the first to be offered new vaccines and indeed, in some but not all cases, will be the volunteers for clinical trials. In 1997, a compulsory anthrax vaccination program was initiated for US military personnel. The vaccines used, the BioT hrax® anthrax vaccine and the Dryvax® vaccinia virus, have both been in use for

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Box 2. National Institute of Allergy and Infectious Disease category B pathogens[101].

- Burkholderia pseudomallei
- Coxiella burnetii (Q fever)
- Brucella species (brucellosis)
- Burkholderia mallei (glanders)
- Ricin toxin (from Ricinus communis)
- Epsilon toxin of Clostridium perfringens
- Staphylococcus enterotoxin B
- Typhus fever (Rickettsia prowazekii)
- Food and waterborne pathogens
- Bacteria
  - Diarrheagenic Escherichia coli
  - Pathogenic vibrios
  - Shigella species
  - Salmonella
  - Listeria monocytogenes
  - Campylobacter jejuni
  - Yersinia enterocolitica
- Viruses (caliciviruses, hepatitis A)
- Protozoa
  - Cryptosporidium parvum
  - Cyclospora cayatanensis
  - Giardia lamblia
  - Entamoeba histolytica
  - Toxoplasma
  - Microsporidia
- Additional viral encephalitides
  - West Nile virus
  - LaCrosse
  - California encephalitis
  - Venezuelan equine encephalomyelitis
  - Eastern equine encephalomyelitis
  - Western equine encephalomyelitis
  - Japanese encephalitis virus
  - Kyasanur Forest virus

preparation as weaponized stocks, and considerable uncertainty as to the precise whereabouts now of all the material from these historic stockpiles. In this sense, despite the need to move towards a response to potential application of any of the agents on the CDC category A–C pathogens list, not all the threats are regarded as equal, and there are more specific concerns over some agents than others. Smallpox is particularly feared for its ease of person-to-person transmission and high incidence of lethality. In the case of smallpox, there is a considerable degree of strategic confidence based on the historic ability of the vaccinia program to eradicate smallpox as a natural pathogen of man.
Box 3. National Institute of Allergy and Infectious Disease category A–C pathogens[101]

- Tickborne hemorrhagic fever viruses
- Crimean–Congo hemorrhagic fever virus
- Tickborne encephalitis viruses
- Yellow fever
- Multidrug-resistant tuberculosis
- Influenza
- Other Rickettsias
- Rabies
- Severe acute respiratory syndrome-associated coronavirus

Emerging infectious disease threats, such as Nipah virus and additional hantaviruses. NIAID priority areas.

more than 50 years, although both have been associated with reports of adverse events. Around 500 members of the US armed forces refused immunizations, a course of events culminating in a legal test case at which Judge Sullivan stated, “Congress has prohibited the administration of investigational drugs to service members without their consent. This court will not permit the government to circumvent this requirement” [10]. This legal precedent will clearly, and quite properly, have far-reaching ramifications for ethical consent issues underpinning development of the forthcoming generation of bioterrorist agent vaccines.

Expert commentary & five-year view

Pulling together the threats, concerns and caveats discussed, it is clear that many hurdles would be overcome if it were possible to move into a new age of high-technology vaccines. The requirements would need to encompass safety, strong potency, but with safe adjuvants such that effective titers could be generated without the need for multiple boosts over long periods, and the incorporation of multiple epitope strings such that defence against several pathogens could be incorporated into a single construct. It is possible that a live vaccine vector could be used to deliver a number of heterologous antigens to achieve multivalent vaccines, or a collection of subunit proteins in an appropriate delivery system could be used to achieve multivalency. An alternative scenario is that such a vaccine construct may need to take the form of a DNA vaccine and that the selected epitopes in the string would need to be selected for antigen presentation and potency across the widest possible range of human leukocyte antigen (HLA)-presenting alleles (9). Although many trials have been undertaken to investigate the immunologic potency of such vaccines in a wide range of disease settings, the field is still in its infancy. A major step was recently taken towards the creation of such vaccines with the establishment, through the NIAID, of an Immune Epitope Data-base and Analysis Resource [2]. However, a number of experimental studies have already been undertaken to investigate the effectiveness of DNA vaccines against biowarfare pathogens. These approaches have included incorporation of anthrax sequences into DNA vaccines, DNA cocktails incorporating both anthrax and plague antigens, or incorporation of anthrax sequences into adenovirus-based vaccine constructs [11–13]. Each approach has given a high degree of protection in animal models.

The development of effective DNA vaccination regimens, coupled with the acquisition of a large database of information on presented epitopes from bioterrorism pathogens, suggests that it may indeed be possible in the near future to progress towards a generation of vaccines that are potent, safe and widely protective.

Key issues

- A major problem exists regarding inability to test the efficacy of candidate vaccines.
- Lessons can be learned from responses to the 2001 release of anthrax spores through the US postal service.
- The list of potential bioterrorist pathogens is wide, although anthrax and smallpox are the major concerns.
- The issue of who to vaccinate and when could be addressed with cost–benefit analysis, but how to go about such an analysis, remains a dilemma.
- The future generation of bioterrorist vaccines will include multi epitope and DNA vaccines - this new age of vaccines is currently in its infancy.

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Website
101 Centers of Disease Control and Prevention www.bt.cdc.gov/agent/agentlist-category.asp (Accessed May 2005)

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