Published in final edited form as:
Future Microbiol. 2009 February ; 4: 35–43. doi:10.2217/17460913.4.1.35.

Anti-toxin antibodies in prophylaxis and treatment of inhalation anthrax

Anette Schneemann† and
Department of Molecular Biology, The Scripps Research Institute, La Jolla, CA 92037, USA, Tel.: +1 858 784 8643; Fax: +1 858 784 7979; aschneem@scripps.edu

Marianne Manchester
Department of Cell Biology, The Scripps Research Institute, La Jolla, CA 92037, USA, Tel.: +1 858 784 8086; Fax: +1 858 784 7979; marim@scripps.edu

Abstract

The CDC recommend 60 days of oral antibiotics combined with a three-dose series of the anthrax vaccine for prophylaxis after potential exposure to aerosolized *Bacillus anthracis* spores. The anthrax vaccine is currently not licensed for anthrax postexposure prophylaxis and has to be made available under an Investigational New Drug protocol. Postexposure prophylaxis based on antibiotics can be problematic in cases where the use of antibiotics is contraindicated. Furthermore, there is a concern that an exposure could involve antibiotic-resistant strains of *B. anthracis*. Availability of alternate treatment modalities that are effective in prophylaxis of inhalation anthrax is therefore highly desirable. A major research focus toward this end has been on passive immunization using polyclonal and monoclonal antibodies against *B. anthracis* toxin components. Since 2001, significant progress has been made in isolation and commercial development of monoclonal and polyclonal antibodies that function as potent neutralizers of anthrax lethal toxin in both a prophylactic and therapeutic setting. Several new products have completed Phase I clinical trials and are slated for addition to the National Strategic Stockpile. These rapid advances were possible because of major funding made available by the US government through programs such as Bioshield and the Biomedical Advanced Research and Development Authority. Continued government funding is critical to support the development of a robust biodefense industry.

Keywords

antibiotic treatment; biodefense funding; inhalation anthrax; lethal factor; medical countermeasures; prophylactic antibodies; protective antigen; vaccination

†Author for correspondence: Department of Molecular Biology, The Scripps Research Institute, La Jolla, CA 92037, USA, Tel.: +1 858 784 8643, Fax: +1 858 784 7979, E-mail: aschneem@scripps.edu.

Financial & competing interests disclosure
Work in A Scheemann and M Manchester's laboratories was supported by grants from the National Institute of Allerg y and Infectious Diseases (P01AI056013 and AI076852). The authors have no other relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript apart from those disclosed.
No writing assistance was utilized in the production of this manuscript.

Anthrax pathogenesis

Bacillus anthracis, the causative agent of anthrax, is a Gram-positive, rod-shaped bacterium that forms highly resistant spores under conditions of environmental stress [1]. Spores represent a dormant, nonreproductive form of the bacterium that is resistant to UV light, desiccation, extreme temperatures and other environmental conditions. Spores can persist in nature for many decades, primarily in soil, and are very difficult to eradicate.

Owing to the soilborne nature of *B. anthracis*, anthrax mainly affects grazing animals but all mammals are susceptible to the disease. Natural infections of humans are rare in the USA and other countries where vaccination of livestock and people who are most likely to come in contact with diseased animals or their products is implemented. In humans, infection is initiated when spores enter the host by one of three routes: the cutaneous route through a cut or abrasion in the skin, the gastrointestinal route by ingestion of contaminated meat and the inhalational route by breathing in airborne spores.

Inhalation anthrax is the deadliest form of the disease because it is difficult to diagnose in a timely manner and since exposure via the inhalational route has the potential to affect a large number of individuals in the event of a deliberate release. Following inhalation, spores are taken up by alveolar macrophages and transported to the mediastinal lymph nodes. During this process, they germinate to form vegetative bacilli that enter the bloodstream and ultimately cause sepsis. The disease has a typical incubation period of 1–6 days and begins with relatively mild, flu-like symptoms such as malaise, fatigue and slightly elevated temperature. This is followed by respiratory distress, which abruptly and rapidly progresses to respiratory failure and shock despite aggressive antibiotic treatment [2]. Two virulence factors, the capsule, which allows the bacterium to evade phagocytosis, and two AB-type exotoxins, lethal toxin and edema toxin, are associated with anthrax pathogenesis [3,4]. The B moiety, protective antigen (PA), represents the cell-binding component required for the entry of the enzymatic A moieties, lethal factor (LF) and edema factor (EF) [5]. LF is a zinc protease that cleaves several mitogen-activated protein kinase kinases leading to blockage of signaling pathways by which immune cells respond to pathogens [6–9]. EF is a calmodulin-dependent adenylate cyclase that generates unphysiologically high levels of cAMP [10]. This leads to impairment of intracellular signaling pathways, interference with phagocytosis by macrophages and disruption of waterhomeostasis with resulting edema [11–13].

Current strategies for prevention & treatment of inhalation anthrax

Prior to the advent of antibiotics, anthrax was treated by passive immunization with animal antisera [2,14]. This practice indicated an important role for antibodies in protecting against the disease. Today there is overwhelming evidence that antibodies are key players in conferring immunity to anthrax. In fact, the protective effect of the anthrax vaccines licensed in the USA (anthrax vaccine adsorbed [AVA]; also known as BioThrax®) and in the UK (anthrax vaccine precipitated) is based on induction of an antibody response to *B. anthracis* proteins, primarily PA. The vaccine is derived from a *B. anthracis* culture supernatant, whose major component is PA with trace amounts of other bacterial components, including EF and LF, which are adsorbed to aluminum hydroxide gel. Numerous studies have confirmed that an antibody response to PA is sufficient to provide protection [15–19]. A major drawback of the AVA vaccine is its lot-to-lot variation, ill-defined general composition and the lengthy course of administration. Six injections over a course of 18 months are considered necessary to induce protection with subsequent annual boosters recommended to maintain immunity. These drawbacks have led to increased efforts in recent years to develop next generation vaccines that are more rigorously defined and confer more rapid protection. The most developed vaccine

candidate is based on recombinant PA expressed and purified from *Escherichia coli* [20] or from an asporogenic, nontoxigenic, nonencapsulated strain of *B. anthracis* [21,22].

Given the short incubation time and rapid disease progression of inhalation anthrax, vaccination is unlikely to afford protection after an individual has been exposed to aerosolized spores. In this situation, antibiotics administered soon after exposure and prior to the onset of symptoms are the most effective means of preventing disease. Since spores can remain dormant in the lungs for an extended period of time [23,24], a 60-day course of oral antibiotics is recommended. This type of prophylactic treatment was effective in the aftermath of the anthrax attacks of 2001, in which close to 10,000 individuals were thought to have been exposed to airborne *B. anthracis* spores and were offered a full course (60 days) of the antibiotics ciprofloxacin or doxycycline. However, a follow-up survey of more than 6000 of these individuals revealed that adherence to the drug regimen was poor. Only 44% of the surveyed individuals followed the prophylaxis protocol correctly whereas others forgot, cited side-effects or stopped because they thought they were not at personal risk [25,26]. The poor compliance is troubling and suggests that additional measures of protection need to be considered in the event of a future mass exposure. Indeed, the most recent CDC recommendations following potential exposure to aerosolized *B. anthracis* spores are 60 days of oral antibiotics combined with a 3-dose series of anthrax vaccine given at 2-week intervals [27]. Because the AVA vaccine is currently not approved by the US FDA for post-exposure prophylaxis, it has to be made available for this purpose under an Investigational New Drug protocol.

Problems associated with postexposure prophylaxis based on antibiotics

Postexposure prophylaxis based on antibiotics can be problematic in cases where use of the recommended antibiotics is contraindicated, for example, in pregnant women and children. A greater concern is the possibility that a future biological attack could involve *B. anthracis* strains that are resistant to antibiotics. Strains naturally resistant to penicillins and cephalosporins have been isolated on occasion [28,29]. In addition, reduced susceptibility as well as complete resistance can be induced in the laboratory by serial passage of *B. anthracis* in the presence of increasing concentrations of numerous other antibiotics [30,31]. Particularly disturbing is the fact that *B. anthracis* strains resistant to the currently recommended antibiotics doxycycline and ciprof loxacin could be generated using straightforward experimental procedures such as transformation of the bacteria with a plasmid containing a tetracycline resistance gene [32] or stepwise adaptation to growth in the presence of high concentrations of ciprofloxacin [31,33]. Finally, antibiotics do not specifically block anthrax toxin action and once significant levels of toxin build up in the bloodstream antibiotic therapy is no longer effective.

Development of immunotherapeutics

Given this background, availability of alternate treatment modalities that are effective in prophylaxis of inhalation anthrax is highly desirable. There has been a major research focus towards passive immunization using polyclonal and monoclonal antibodies (mAbs) against *B. anthracis* toxin components, primarily PA and to a lesser extent LF. These antibodies would provide immediate and extended protection against lethal toxin (LeTx) given the relatively long half-life of immunoglobulins in serum. Results that have emerged from numerous investigations strongly support the notion that anti-PA and anti-LF antibodies are beneficial in pre- and postexposure prophylaxis.

One of the first studies to indicate that anti-PA antibodies offer protection in the absence of antibodies against other *B. anthracis* components, employed polyclonal antisera from guinea pigs that had been vaccinated with recombinant PA [16]. Passive administration of this

antiserum to other guinea pigs that were challenged intramuscularly with a lethal dose of *B. anthracis* Ames spores protected 67% of the animals. This level of protection was comparable to that achieved by active immunization of guinea pigs with AVA. In a subsequent study it was demonstrated that purified rabbit and sheep anti-PA IgG protected 90–100% of mice challenged with a lethal dose of *B. anthracis* Sterne spores when the antibodies were administered in combination with ciprofloxacin. By contrast, treatment with antibodies or antibiotics alone protected only 30–50% of the animals [34]. These and other preliminary studies [35,36] laid the foundation for more systematic and rigorous follow-up investigations, most of which focused on characterizing the protective capacity of monoclonal anti-PA and anti-LF antibodies. mAbs offer several advantages over polyclonal antibodies, including defined specificity, reproducible affinity, high purity and increased safety. In addition, mAbs can be engineered to eliminate effector function, alter serum half-life or enhance activity through bioconjugation of drugs, toxins and radioisotopes.

Murine mAbs

The fact that murine mAbs against PA or LF have the capacity to neutralize LeTx was demonstrated early on by *in vitro* cell intoxication assays and *in vivo* LeTx challenge of Fisher 344 rats [37,38]. Subsequent studies confirmed that anti-PA and anti-LF mAbs also protect animals from lethal spore challenge [39]. Mohamed *et al.* reported an unexpected result as they observed that certain mAbs against PA enhance rather than neutralize LeTx cytotoxicity, apparently in a Fc receptor-dependent manner [40].

Humanized mAbs

Studies that are of particular interest in this area of research are those that have led to the isolation of human or humanized mAbs against PA and LF. Animal-derived mAbs are ultimately not suitable for clinical applications given the reactivity against nonhuman proteins, especially when they are administered repeatedly. The approaches taken to develop human mAbs are diverse and highlight the advances made in the antibody engineering field in recent years.

Several humanized mAbs with very high affinities for PA were generated by recombinant phage technology using a variety of strategies. Wild *et al.* employed phage libraries displaying Fab fragments derived from lymphocytes of AVA-vaccinated donors and subjected them to several rounds of panning against trypsin-cleaved PA fragment (PA63) in the presence of full-length PA [41,42]. Of several *in vitro* neutralizing Fabs that were isolated, further studies with two Fabs, 63L1D and 83K7C, with affinities to PA63 of K_d 0.13 and 0.87 nM, respectively, showed protection against LeTx challenge in rats. Fab 63L1D appeared to neutralize by blocking the binding of LF to PA63. Conversion of the Fab fragments to full-length human IgG1 molecules allowed an approximate sevenfold reduction in the amount of antibody required to neutralize the toxin *in vivo*. Maynard *et al.* constructed single-chain variable fragments (scFvs) from mAb 14B7, which inhibits binding of PA to the cell receptor [37,43], and used error-prone PCR to prepare affinity-enhanced clone 1H scFv, which had a K_d of 0.25 nM. Clone 1H scFv neutralized LeTx cytotoxicity *in vitro* with a lower IC_{50} than mAb 14B7 and provided better protection *in vivo* against a LeTx challenge in rats. Mohamed *et al.* prepared an affinity-enhanced chimeric, de-immunized mAb ETI-204, by fusing the 14B7 V_H and V_L genes to human Ig constant regions [44]. ETI-204 retained high affinity for PA (K_d = 0.33 nM) and protected 90–100% of rabbits when administered intravenously and 100% of rabbits when administered intramuscularly before aerosol challenge with *B. anthracis* Ames spores. Partial protection was observed in rabbits injected intravenously at 24 (8/10), 36 (5/10) and 48 h (3/7) after aerosol challenge with *B. anthracis* Ames spores [44]. Rabbits surviving challenge from one of the experiments also developed anti-PA titers [44]. The intramuscular route of antibody

administration is favorable compared with the intravenous route as it facilitates treatment of large numbers of people in case of a mass attack.

Human mAbs

A strategy for the isolation of intact human monoclonal anti-PA antibodies used by Sawada-Hirai *et al.* entailed collection of peripheral blood lymphocytes from AVA-immunized donors and subsequent transplantation of the lymphocytes into severe combined immunodeficient mice [45]. Challenge of the mice with PA and LF led to the proliferation of PA-specific plasma cells which were isolated and immortalized by hybridoma formation. Since human/murine hybridoma cell lines tend to be unstable, the genes encoding the antibodies were cloned and transferred to Chinese hamster ovary cells for stable expression of full-length human immunoglobulin. One of several anti-PA antibodies isolated by this method, AVP-21D9, had an exceptionally high affinity for PA (K_D = 82 pM) and fully protected rats from LeTx challenge when administered as early as 17 h prior to LeTx infusion. Even 1 week after antibody administration, 80% of rats were still protected from LeTx challenge when a tenfold higher dose of antibody was used. Molecular analyses showed that AVP-21D9 inhibits PA heptamer formation, a step that is critical for binding EF and LF prior to entry into susceptible cells [46]. Subsequent experiments by Peterson *et al.* confirmed that AVP-21D9 fully protects mice from lethal challenge with *B. anthracis* Ames spores by the intranasal route as long as the antibody is administered within 24 h of infection and provided that the animals receive a daily, albeit suboptimal, dose of ciprofloxacin [18]. Antibody alone or a suboptimal dose of ciprofloxacin alone did not confer full protection. Rechallenge of the surviving mice with another lethal dose of *B. anthracis* Ames spores resulted in death of all animals indicating that antibody plus antibiotic treatment does not lead to the induction of a protective immune response in mice [18]. Similar results were obtained with guinea pigs, whilst in contrast, rabbits were fully protected from a lethal spore challenge by AVP-21D9 in the absence of ciprofloxacin even when the antibody was administered 12 h after spore challenge. Moreover, the rabbits were protected when challenged subsequently with another lethal dose of spores [18,47]. These studies emphasize that small animal models of inhalation anthrax differ significantly in their response to passive immunization and that rabbits are more easily protected than mice and guinea pigs.

In an alternate approach, Vitale *et al.* injected HuMab transgenic mice [48] that were engineered to express human immunoglobulins [49] with recombinant PA and screened for *in vitro* neutralizing activity. This strategy led to the isolation of several mAbs, one of which, mAb1303, was selected for further studies based on its high potency in *in vitro* toxin neutralization assays. Purified mAb1303 was shown to have prophylactic activity in the rabbit model of anthrax inhalation. Specifically, rabbits were protected from lethal aerosol challenge with Ames spores when mAb1303 was administered intravenously 1 h and 3 days after exposure. Similarly, mAb1303 conferred complete protection in nonhuman primates when administered intramuscularly 1 h after spore challenge. mAb1303 was also therapeutically active as demonstrated by the protection of rabbits that received the antibody either 24 or 48 h after aerosol challenge. Interestingly, the ability of mAb1303 to neutralize LeTx not only depends on its interaction with PA but also on interaction with Fc receptors [48]. Fab fragments of mAb1303 did not neutralize LeTx and full-length antibody was not capable of LeTx neutralization when Fc receptors were blocked. The mechanism underlying toxin neutralization by mAb1303 antibody is not yet fully understood. This result is particularly remarkable given that other anti-PA mAbs were shown to enhance LeTx toxicity in a Fc receptor-dependent fashion [40].

Commercial development of human & humanized mAbs

Several of the human or humanized anti-PA mAbs described are being commercially developed for application in pre- and postexposure inhalation anthrax as well as for therapeutic treatment of the disease. Avanir Pharmaceuticals (CA, USA) initially isolated and characterized AVP-21D9 but subsequently sold the rights for further development to Emergent Biosolutions (MD, USA), the manufacturer of the anthrax vaccine AVA. AVP-21D9 has yet to be tested in nonhuman primates before it can advance to clinical studies.

Elusys Therapeutics (NJ, USA) is developing the humanized, affinity-enhanced mAb ETI204 under the tradename 'Anthim™', for pre- and postexposure prophylaxis of inhalation anthrax as well as for treatment of the disease. In a Phase I clinical study, Anthim was shown to be safe and well tolerated when given to healthy volunteers at the anticipated therapeutic dose with or without ciprofloxacin. The drug has received Fast-Track and Orphan Drug status by the US FDA and is being developed for intramuscular delivery using prefilled syringes or autoinjectors.

PharmAthene Inc. (MD, USA) in collaboration with Medarex (NJ, USA) is developing the fully human mAb1303 under the tradename Valortim® for inhalation anthrax prophylaxis and therapy. A Phase I clinical trial has been completed showing that administration of Valortim by the intravenous and intramuscular route is safe and well tolerated. Like Anthim, Valortim has received Fast-Track and Orphan Drug status by the FDA.

Another human mAb isolated by Human Genome Sciences (MD, USA), PamAb (Abthrax™), has also completed Phase I clinical trials [50]. Abthrax functions by inhibiting binding of PA to its receptor. A single dose of this antibody increased survival rates of monkeys from inhalation anthrax by 64%, even when administered after animals showed signs of the disease [101]. In Phase I trials it was shown that the antibody is safe, well-tolerated and bio-available after a single intramuscular or intravenous dose [50]. Co-administration of the antibiotic ciprofloxacin did not affect pharmacokinetics of either drug, indicating that they can be used in combination for prophylaxis and treatment of inhalation anthrax [101]. Human Genome Sciences is currently manufacturing Abthrax to begin delivery of 20,000 doses to the National Strategic Stockpile in 2009.

Human polyclonal antibodies

Although monoclonal anti-PA antibodies represent a welcome addition to the arsenal of prophylaxis and treatment options for inhalation anthrax, their monospecific nature makes it possible to develop *B. anthracis* strains that resist their action. By mutating the epitope to which the antibody binds, the drug will lose its effectiveness, while the functions of PA, such as receptor binding, heptamer formation and endocytosis to deliver LF and EF, may not be affected. This scenario could be avoided if cocktails of mAbs were employed that target different PA epitopes or that include mAbs recognizing additional *B. anthracis* components such as LF and capsule. The isolation of anti-LF mAbs and anticapsule mAbs, which are effective in neutralizing LeTx *in vivo* and *in vitro*, has been reported by several groups [51–55]. A fully human mAb against LF is being commercialized under the name Anthraxumab by IQ Corporation (The Netherlands) [102].

Meanwhile, Cangene Corporation (Winnipeg, Canada) [103] and Emergent Biosolutions [104] both manufacture polyclonal immunoglobulin or 'Anthrax Immune Globulin' (AIG) from plasma of human volunteers who have been vaccinated with AVA. The advantage of AIG is that its antibody composition reflects the breadth of the natural immune response and therefore offers the possibility of a more efficient induction of effector mechanisms such as complement-induced bacterial cell lysis, antibody-dependent cellular cytotoxicity,

phagocytosis, and so on. There are also clear disadvantages, such as the limited availability of donor blood, batch-to-batch variation, the risk of infectious disease transmission and the high cost of production. These issues could be circumvented by taking advantage of recent advances in the production of recombinant polyclonal antibodies [56]. The US government has announced plans to purchase 10,000 doses of AIG manufactured by Cangene for the National Strategic Stockpile.

AIG was recently used under an Emergency Investigational New Drug use protocol in a patient who had naturally acquired inhalation anthrax [57]. The patient presented to a local hospital with symptoms of mild respiratory distress and initially received aggressive antibiotic treatment as well as other critical support. When the patient's condition deteriorated, AIG was added to the treatment protocol based on a recommendation by the CDC. LF in serum plasma and pleural fluid dropped sharply after administration of AIG, suggesting that it had a beneficial effect. However, more systematic and controlled studies are necessary to confirm that this response was due to the infusion of anthrax immunoglobulins. The patient eventually recovered.

Alternate anti-toxin countermeasures

Antibodies are not the only reagents expected to be useful in prophylaxis and treatment of inhalation anthrax. In principle, any compound capable of neutralizing anthrax toxins represents a possible alternative. One approach that is currently pursued entails the use of anthrax toxin receptor, CMG2, as a toxin antidote. Soluble CMG2 was shown to inhibit LeTx *in vitro* and *in vivo* based on competition with cellular CMG2 for PA binding [58]. The use of soluble CMG2 as an anti-toxin has the distinct advantage that *B. anthracis* strains resistant to its action would be difficult, if not impossible, to engineer since any mutation in PA preventing interaction with soluble CMG2 would also prevent its binding to the cell. An alternate means of exploiting the anti-toxin function of CMG2 is multivalent display of CMG2 on a viral platform. This form of CMG2 was shown to function not only as an inhibitor of LeTx but also as a potent vaccine in the PA-bound form [59]. The vaccine afforded protection from LeTx challenge in rats within 3 weeks after a single injection. Another strategy under investigation involves the use of a PA derivative that functions as a dominant inhibitor of PA heptamer formation [60,61], thereby interfering with LF and EF binding and cell entry. Equally important are efforts currently underway to identify potent small molecule inhibitors of the enzymatic activities of LF and EF. Small molecule inhibitors would provide a cheaper alternative to protein-based reagents and might be orally available.

Future perspective

The recent advances in basic and applied research of medical countermeasures against inhalation anthrax have been remarkable. Since 2001, the US government has made more than US$40 billion available to accelerate biodefense research and development through programs such as Bioshield and The Biomedical Advanced Research and Development Authority. These funds have enabled small biotech firms to design and develop novel products for the prevention and treatment of diseases resulting from potential biowarfare with a variety of agents. The further commercial development, procurement and stockpiling of these countermeasures, however, has not been supported as aggressively and the industry cannot develop financial robustness in response to the needs of a single government customer. This situation has created unique challenges that result from a combination of factors including the currently unpredictable market for and profitability of potential products, leading to limited interest from private investors. Several small biotech companies have spearheaded the effort to turn biodefense research into a commercial enterprise, but to date large pharmaceutical companies have not been attracted to this endeavor. Long-term investment in biodefense research,

development and procurement by the government is critical to sustain this young industry and vital for maintaining an effective defense against biological pathogens.

Executive summary

Current strategy for prophylaxis of inhalation anthrax

- Antibodies to *Bacillus anthracis* proteins, in particular protective antigen (PA), confer immunity to inhalation anthrax and can be induced by vaccination with anthrax vaccine adsorbed (AVA) or anthrax vaccine precipitated. Vaccination is unlikely to afford protection when implemented after an individual has been exposed to aerosolized *B. anthracis* spores given the short incubation time and rapid progression of the disease. In this situation, prophylactic treatment with antibiotics is effective when initiated prior to the onset of symptoms and if maintained over a period of at least 60 days.

Problems associated with the existing strategy

- Following the 2001 anthrax attacks, surveys showed that fewer than half of the individuals potentially exposed to *B. anthracis* spores correctly followed the antibiotic treatment protocol suggesting that additional measures of protection need to be considered. Moreover, antibiotic treatment is contraindicated in certain cases. Of great concern is the possibility that a future attack involves *B. anthracis* strains resistant to antibiotics.

Development of alternate or supplemental prophylaxis strategies

- A need exists for alternate or supplemental treatment modalities, particularly those that neutralize anthrax toxins. The focus has been on passive immunization with monoclonal and polyclonal antibodies to *B. anthracis* toxin components, primarily PA and to a lesser extent lethal factor (LF).
- Humanized and fully human monoclonal antibodies (mAbs) with high affinities for PA and LF have been generated using a variety of approaches. These mAbs afford significant protection from inhalation anthrax in small animal models and nonhuman primates in a prophylactic and therapeutic setting. Several of these antibodies have passed Phase I clinical trials. Abthrax™, an anti-PA mAb developed and manufactured by Human Genome Sciences (MD, USA), will be added to the National Strategic Stockpile in the fall of 2008.
- A potential disadvantage of mAbs is their monospecific nature, which makes it possible, in principle, to develop *B. anthracis* strains that resist their action. A cocktail of mAbs that target multiple epitopes of one protein or more than one *B. anthracis* toxin component would be preferable.
- Polyclonal immunoglobulin, ‘Anthrax Immune Globulin’, from the plasma of human AVA-vaccinated volunteers, is manufactured as an alternative to mAb preparations. The advantage of Anthrax Immune Globulin is that it reflects the breadth of the human immune response to *B. anthracis*; however, there is limited availability, lot-to-lot variation and it carries the risk of infectious disease transmission. The development of recombinant human polyclonal antibody preparations should be considered.
- Other anthrax anti-toxins are in research phase with the main focus on soluble or conjugated forms of CMG2 as a lethal toxin antidote and a dominant-negative form of PA that interferes with heptamer formation.

- Small-molecule inhibitors of the enzymatic activities of LF and edema factor are highly desirable given their potential oral availability and low cost of production
- The commercial development of medical countermeasures for biodefense is facing unique challenges resulting, in part, from small market for potential products, low profit margins and limited private investor interest. The biodefense industry, made up of several small biotechnology firms, relies heavily on funds made available by the US government.

Summary

- Antibodies that neutralize anthrax toxins have been developed and shown to afford protection from inhalation anthrax in a prophylactic and therapeutic setting. These antibodies are likely to be used in combination with antibiotics. The development of new medical countermeasures against inhalation anthrax and other diseases caused by biological warfare agents has been spearheaded by small biotech companies. These companies depend on continued government funding to maintain an active biodefense research and development program.

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