Department of Health and Human Services
Public Health Service
Food and Drug Administration
Center for Biologics Evaluation and Research

To: File for STN 125562

From: Robert W. Fisher, DHRR, OBRR, CBER

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Applicant: Cangene Corporation

Product: Anthrax Immune Globulin Intravenous (Human), AIGIV; Anthrasil™

Subject: Final review, STN 125562/0 non-human primate studies and Essential Data Elements for an Animal Model

Recommendation:

      Approval.

Executive Summary:

      Cangene Corporation (Cangene) has submitted a Biologics License Application for Anthrax Immune Globulin Intravenous (Human) [AIGIV] with an indication for the treatment of inhalational anthrax in adults and pediatric patients in combination with appropriate antibacterial drugs. Cangene's submission was found to be consistent with the 'Essential Data Elements of an Animal Model' described in the 2009 "Draft Guidance for Industry: Animal Models- Essential Elements to Address Efficacy Under the Animal Rule". The efficacy of AIGIV has been demonstrated in two animal models: the cynomolgus macaque (*Macaca fascicularis*) inhalational anthrax challenge model and the New Zealand White rabbit inhalational anthrax challenge model. The model development and efficacy data from the nonhuman primate model indicate that AIGIV is reasonably likely to provide clinical benefit in humans with inhalational anthrax.

Background:

1. STN 125562/0 is an eCTD format original Biologics License Application (BLA) for Anthrax Immune Globulin Intravenous (Human) [AIGIV] submitted by Cangene Corporation.

a. This submission was received at DCC on 25 July 2014 and a chair assigned on 28 July 2014.

b. AIGIV was developed under Cangene's BB-IND 11982, received at FDA on 12 October 2004.

    i. Fast Track designation for AIGIV development was granted on 21 December 2006.

    ii. Orphan drug status was granted for 'treatment of toxemia associated with inhalational anthrax' on 29 July 2008. The orphan drug indication is being modified to 'treatment of inhalational anthrax' subsequent to FDA's request to simplify AIGIV's indication.

    iii. The Strategic National Stockpile began acquiring AIGIV in March 2006 and currently has accepted (b) (4) lots into inventory.

    iv. A pre-emergency use authorization (pre-EUA) (submission ID 14456) from the Centers for Disease Control and Prevention (CDC) for use of AIGIV in the event of a declared emergency was acknowledged on 10 August 2010.

    v. CDC has an IND (BB-IND 13026) for use of AIGIV to treat sporadic cases of anthrax, however prior to March 2010 individual emergency INDs were required for actual use of the product.

        1. AIGIV was used under INDs 12953, 13867, 14249, 14246, 12427, 14261, 14272, 14270, 14288, 14297, and 14301.

        2. Health Protection Scotland (HPS) purchased three doses of AIGIV directly from Cangene for use in treating patients with injectional anthrax. This was not captured in a US IND, however HPS utilized a CDC protocol (IRB 4881, version 5.0, dated 18 December 2009) as the reference treatment protocol.

c. Priority review was discussed in the 13 February 2014 responses to the preBLA meeting request (IND 11982/191).

    i. Priority review was approved as communicated to the sponsor in the filing letter dated 23 September 2014.

d. The Action Due Date for this BLA is 25 March 2015.

2. AIGIV is a polyclonal antibody preparation manufactured from the plasma of humans immunized with Anthrax Vaccine Adsorbed (AVA; Biothrax).

a. AVA is a vaccine prepared from cell-free filtrates of a nonencapsulated, avirulent strain of *Bacillus anthracis* (strain V770-NP1-R). Protective antigen (PA), a 83kDa protein, is known to be an immunodominant component in this type of cell-free vaccine [1], however numerous other immunogenic proteins are present [2,3] and antibodies to the anthrax lethal factor (LF) and edema factor (EF) proteins have been found in vaccines [4,5].

b. The antibody response induced by AVA can confer immunity to anthrax in humans [6], and protection has been attributed to PA neutralizing antibodies in animal models [7–9].

c. Monoclonal antibodies against PA can also provide therapeutic benefit in animal models of anthrax [10]; one such monoclonal (raxibacumab) was approved in 2012 for the treatment of inhalational anthrax based on animal efficacy data.

d. Potency of the AIGIV product is determined via a cell based assay that measures the capacity of the product to protect cells from B. anthracis lethal toxin (LT; composed of PA and LF) induced cytotoxicity. An in-house reference standard is used with assigned units

where (b) (4) of standard (b) (4) is equivalent to (b) (4) anti-PA activity.

   e. AIGIV is produced using Cangene's column-based immunoglobulin purification platform. The product has a minimum potency of 60 Units/vial.

3. Anthrax is a rare illness triggered by infection with *B. anthracis*, a gram-positive, encapsulated, spore-producing bacterium. There are multiple forms of the disease, dependent upon the route of exposure: inhalational, gastrointestinal, cutaneous, and (more recently described) injectional. Anthrax is a CDC Category A pathogen and is considered a Tier I overlap select agent by the U.S. Department of Health and Human Services and the U.S. Department of Agriculture primarily due to the risk from aerosolized anthrax spores. The spores are extremely hardy and their ~1µm diameter is ideal for deposition in the lungs if inhaled. Incidence in the United States is exceedingly low, with 1-2 cases per year (primarily the cutaneous form) however a spike in inhalational cases was noted in 2001 as a result of intentional dissemination of finely divided anthrax spores in the U.S. mail. This attack emphasized the risk from the deliberate use of anthrax, a known bioweapon that was placed into production status by the U.S. and the U.S.S.R. [11–13].

   a. Inhalational anthrax is triggered when *B. anthracis* spores are inhaled and deposited in the lung.

      i. The spores are phagocytized by alveolar macrophages that traffic to regional lymph nodes. The spores germinate within the phagosomes of infected macrophages, and the vegetative bacteria produce Lethal Factor (LF), Edema Factor (EF) and PA[14]. During anthrax infection, PA binds to one of two cellular receptors (tumor endothelial marker 8 or capillary morphogenesis gene 2) on host cells. A furin cleavage event activates PA and triggers the formation of a PA heptamer that can bind LF or EF; PA complexed with EF or LF forms edema toxin (ET) and lethal toxin (LT), respectively. These complexes undergo endocytosis and the internalized ET and LT interfere with critical cellular pathways[15]. ET is an adenylyl cyclase, which increases intracellular cAMP levels, and triggers influx of interstitial fluid, resulting in edema. LT inactivates mitogen-activated protein kinase kinases and interferes with the host immune response by triggering apoptosis of macrophages. The bacteria disseminate widely, continue replicating, and contribute to hemorrhagic mediastinitis, septicemia, meningitis, and eventually death. The concentration of circulating PA correlates with the extent of bacteremia in the blood of experimentally infected animals[16] and is a useful marker of infection.

      ii. The incubation period following aerosol exposure of *B. anthracis* spores ranges from one to 43 days, and the disease follows a biphasic clinical course. The initial symptoms are often nonspecific. Malaise, fever, diaphoresis, cough, chest discomfort, nausea, and vomiting were common in the 2001 attack patients, and these patients sought care after a median of 3.5 days after symptom onset[17]. In the absence of treatment, and sometimes despite treatment, the disease becomes fulminant and progresses rapidly; this state is characterized by hypotension, dyspnea, cyanosis, respiratory failure (often resulting from massive pleural effusions), and shock; this fulminant stage may be preceded by a period of apparent improvement in constitutional symptoms. Pleural effusion is common and may require aggressive drainage to maintain lung function as well as to remove

a large potential toxin reservoir[18,19]. Anthrax meningitis can complicate inhalational and other forms of systemic anthrax and has an extremely high mortality rate despite aggressive therapy.

      iii. Inhalational anthrax in general is highly lethal. Prior to 1976 the case fatality rate in the United States was 88% (16/18)[20]. During the Sverdlovsk incident, where an accident at a Soviet biological warfare production facility lead to release of 10^9-10^{12} respirable spores (composed of a mass of between 0.25 and 2.3 grams)[21], approximately 86% (66/77) of reported human cases died and livestock deaths were reported up to 50 km downwind [22]. The prognosis was improved for patients from the 2001 bioterrorism attack where 46%[17] (5/11) died, perhaps due to use of multiple antibiotic treatments and improved supportive care. Survival is rare in untreated cases[23].

      iv. The estimated mean lethal dose of anthrax spores for humans via inhalation is ~1×10^4 (ranges vary between 8×10^3-5.5×10^4 spores), assuming that the disease is uniformly lethal in untreated individuals.[21 24]

b. Gastrointestinal anthrax includes both oropharyngeal and intestinal types and is often linked to consumption of anthrax-contaminated meat. The former presents with ulcers in the mouth and throat, sore throat, dysphagia, regional lymphadenopathy, and swelling of the neck. A 1982 outbreak of anthrax in Thailand yielded 24 cases of oropharyngeal anthrax and three deaths (12.5%) despite antibiotic treatment in all 24 patients[25]. Intestinal anthrax is caused by anthrax infection of the stomach and/or intestine and presents with nonspecific symptoms including nausea, vomiting, abdominal pain, and diarrhea. Ulceration usually occurs in the bowel and, like the other forms of anthrax, may result in bacteremia and sepsis. Patients with a rapid onset of symptoms (<48h) tend to fare poorly despite antibiotic treatment[26].

c. Injectional anthrax is a relatively modern manifestation of anthrax infection in soft tissue, first described in 2001 in a heroin user[27]. This anthrax type results from injecting drugs contaminated with anthrax spores. There is extensive local edema and skin changes consistent with drug use; surgical debridement may be necessary to remove damaged tissue. Notably an eschar does not form, and without clinical suspicion of illicit drug use injectional anthrax may be difficult to detect early. An outbreak in the United Kingdom and Europe in 2009-2010 resulted in 52 cases with 18 deaths[28].

d. Cutaneous anthrax is typically less lethal than the other forms of the disease in part due to the distinct presentation and resulting likelihood for early antibiotic treatment. This form of anthrax results when spores are introduced through breaks in the skin and germinate. Severe local edema, ulceration, and the formation of a black, necrotic eschar that sloughs after 1-2 weeks follow. As with the other forms of anthrax, bacteremia and toxemia may develop in unrecognized or untreated cases and the prognosis in this situation is poor[29].

4. Cangene submitted final study reports for 5 nonhuman primate (NHP) studies. This total includes the initial model development study ((b) (4) 711-G5780), a pharmacokinetic study in unchallenged NHP ((b) (4) 695-G5780), a delayed time-course ciprofloxacin efficacy study ((b) (4) FY09-025), an evaluation of AIGIV therapeutic efficacy in combination with ciprofloxacin ((b) (4) 987-G5780) and the pivotal AIGIV efficacy study ((b) (4) 828-G5780).

5. Study 711-G5780 was the initial model development study performed in NHPs.

    a. This was a Good Laboratories Practice (GLP; per 21 CFR Part 58) study designed to examine the physiological response of cynomolgus macaques (*Macaca fascicularis*) exposed to an aerosol of *B. anthracis* Ames spores. The study was initiated 07 April 2008, and was performed at (b) (4)

    b. A cohort of 12 NHP (6 male and 6 female) was anesthetized with Telazol and exposed to a target (planned) dose of 200 LD_{50} inhaled spores (1.2×10^7 colony forming units, cfu). The actual dose was 240 +/- 51 LD_{50}, with a mass median aerodynamic diameter of 1.1 µm. A group of 4 NHP (2 male, 2 female) was likewise anesthetized but was not exposed to the challenge agent and served as uninfected controls.

    c. Body temperature and activity was monitored hourly via telemetry implant, and physical signs were monitored every 6 hours on study days 1-10 and every 12 hours on study days 11-28. Body weights were monitored on study days 0-10 and on days 14, 21, and 28. Blood was collected via a vascular access port on study day -7, at 24, 30, 36, 42, 48, 54, 60, 66, and 72 hours post-challenge, and daily on study days 4-6.

        i. Hematology tests include WBC, differential leukocyte count, HGB, HCT, RBC, MCV, MCH, MCHC, RDW, PLT, MPV, and N/L ratio. C-reactive protein levels were monitored, and bacteremia was evaluated with (b) (4) . Toxemia was examined via an (b) (4) specific for *B. anthracis* protective antigen.

    d. All anthrax-challenged animals were found dead (n=7) or were euthanized (n=5) based on the pre-specified euthanasia criteria. No unchallenged animals died or exhibited any signs of illness. Mean time to death in the anthrax challenge group was 109 hours, with a median survival of 98 hours. Lethargy and inappetence were the most frequently observed clinical signs. Infected animals demonstrated PA levels greater than the lower limit of detection of the assay ((b) (4)) by 36 hours, and all challenged animals were positive by 48 hours; mean time to a positive PA result was 41.38 hours with a 95% confidence interval of 37.7-45.1 hours. ECL correlated significantly with positive bacteremia status (qualitative p=0.0060, quantitative p=0.0008) but not time to death (p=0.0675). All 12 challenge group animals developed bacteremia. Mean time to positive bacteremia status by direct culture (qualitative) was 42.5 hours, with a 95% confidence interval of 39.5-45.5 hours. Mean time until positive (b) (4)) was 48.9 hours (95% confidence interval of 43.3-54.5 hours). Other study parameters (temperature, CRP, N/L ratio) were variable but did serve to indicate disease progression. 10/12 animals demonstrated three consecutive increases in temperature above baseline (two animals were excluded due to telemetry errors per DR-5269) with a mean time of 36.8 hours.

    e. In conclusion study 771-G5780 was adequately conducted and provided reasonable evidence that bacteremia and toxemia provide evidence of anthrax disease in this animal model. For details refer to the review memo for BB-IND 11982 amendment 95, dated 17 August 2011.

6. Study (b) (4) 695-G5780 was a single dose PK study in NHPs.

    a. This was a GLP study to evaluate the pharmacokinetics of AIGIV lot 10602912 following a single intravenous infusion in uninfected NHPs. Two doses of AIGIV were evaluated: 5

U/kg, and 30 U/kg.

    i. This study was one of the two NHP studies audited by BIMO. The inspection was classified as Voluntary Action Indicated.

b. For details refer to Dr. Mahmood's clinical pharmacology memorandum dated 17 December 2014 as amended. Briefly, based on TNA assay results the AUC_∞ in the 5 U/kg dose group (n=1 due to other animals failing PK modeling acceptance criteria) was 752 day*mU/mL. The AUC_∞ in the 30 U/kg dose group (n=7) was 2490 day*mU/mL. C_{max} was 104 mU/mL and 533 mU/mL and clearance was 6.55 mL/day/kg and 12.5 mL/day/kg in the 5 U/kg and 30 U/kg dose groups, respectively; see Table 1, below. There was no significant impact of sex on the PK parameters of AIGIV in this study.

    i. For comparative purposes, please note that the Cangene proposed dose of 420 U in humans resulted in an AUC_∞ of 2507 day*mU/mL in normal human volunteers (study AX-001), and a 30 U/kg dose of AIGIV resulted in a 2450 day*mU/mL AUC_∞ in rabbits. At the 30 U/kg dose, however, Cmax was ~3.5x higher in the animals compared to the human 420 U dose.

    ii. Based on AUC, it may reasonably be concluded that the 420 U dose in humans is equivalent to a 30 U/kg dose in either rabbits or cynomolgus macaques. However, one potential caveat is that the rabbits and NHPs used in the PK studies were of similar weights (large rabbits, small NHPs) so that dose extrapolation may be confounded by weight as well as lack of robust data in the 5 U/kg NHP group.

7. Study (b) (4) FY09-025 was a randomized, non-GLP study to determine the time at which ciprofloxacin treatment would result in reduced protection in cynomolgus macaques with inhalational anthrax. This information was necessary to evaluate any potential added benefit of AIGIV in combination with antibiotics in the nonhuman primate model.

a. Forty-two NHPs (cynomolgus macaques of Vietnamese origin) were implanted with vascular access ports for blood sampling and randomized to five gender-balanced groups.

b. The NHPs were exposed to aerosolized *B. anthracis* Ames spores at a target dose of 200 LD_{50}.

c. Group 1 (n=6) was designated as the control group and received a single gavage of water 48 hours post challenge. Group 2 (n=8) received an oral loading dose of 32 mg/kg 48 hours post challenge and 16 mg/kg ciprofloxacin orally every twelve hours thereafter for a total of 10 treatments. Group 3 (n=8), group 4 (n=10), and group 5 (n=10) were treated in an identical manner with treatment starting at 60, 72, and 84 hours post-challenge, respectively. Ciprofloxacin was administered via naso-esophageal or oro-esophageal gavage.

d. The average anthrax spore dose was 194 LD_{50}, with a range of 162-205 LD_{50}, and the challenge aerosols had a mean aerodynamic diameter of 1-3 µm. The anthrax spore challenge dose was not statistically different across the five treatment groups.

e. Two animals, A07351 (Group 1) and A06661 (Group 2) were excluded from the survival analysis due to death from non-anthrax causes (systemic *Staphylococcus aureus* infection). Interestingly this strain of *S. aureus* was apparently resistant to ciprofloxacin since A06661 died on study day 7 despite 5 days of ciprofloxacin treatment.

    i. 26/36 animals in the ciprofloxacin treatment groups died before completing the full

antibiotic regimen. Of these, 4 animals (1 animal in group 4, 3 animals in group 5) died before receiving the initial ciprofloxacin dose.

        ii. No animals in group 1 survived (control) compared to 71% (2/7) in group 2, 38% (3/8) in group 3, and 10% (1/10) in each of groups 4 and 5. Only group 2 was significantly different from group 1 based on a logrank test. See Figure 1 and Table 2.

8. Study (b) (4) 987-G005780 was a randomized, non-GLP study to evaluate AIGIV efficacy in NHPs (cynomolgus macaques) when administered in combination with antibiotic therapy after an anthrax aerosol challenge.

    a. Three groups of 20 NHPs (10 male, 10 female) and one group of 12 NHPs (6 males, 6 females) were exposed to an anthrax challenge dose averaging 366 ± 115 LD$_{50}$ and a mass median aerodynamic diameter of ~ 1.3 µm.

    b. Group 1 animals (n=12) were untreated, and Group 2-4 animals (n=20 each group) received oral ciprofloxacin twice daily for 5 days starting at 64 hours post-challenge. Ciprofloxacin treatment involved a loading dose of 32 mg/kg followed by maintenance doses of 16 mg/kg. In addition, Group 2-4 animals received IGIV placebo (Group 2), 15 U/kg AIGIV (Group 3), or 30 U/kg AIGIV (Group 4) in conjunction with the ciprofloxacin loading dose.

    c. Survival at 28 days was evaluated in NHPs with confirmed anthrax infection. Not all animals survived to the 64 hour timepoint to receive antibiotic treatment; the cause of death for 4 animals could not be positively attributed to anthrax.

        i. Overall 93% of the NHPs were positive for PA, including all of Group 1 and all of Group 3.   Including only animals bacteremic prior to treatment, survival in the control group (Group 1) was 8% (1/12). The lone survivor in this group had a single positive result for bacteremia at day 4 and did develop toxemia 30 hours post-challenge, which resolved by day 10 suggesting some level of pre-existing immunity. Survival in the IGIV + ciprofloxacin arm (Group 2) was 75% (9/12). The 15 U/kg AIGIV + ciprofloxacin group (Group 3) and 30 U/kg AIGIV + ciprofloxacin group (Group 4) exhibited 83% (10/12) and 79% (11/14) survival, respectively. Survival in all antibiotic treatment groups (Groups 2-4) was statistically significant compared to the untreated arm (Group 1), but there was no significant difference between Groups 2-4. Toxemia (as measured by PA levels) was decreased in Groups 3 and 4 compared to Groups 1 and 2. See Table 3. The conclusion of this study was that 1) no added benefit had been demonstrated however interference could be ruled out, and 2) additional added benefit studies in the nonhuman primate model of inhalational anthrax would be counterproductive due to the very large study size required to achieve sufficient statistical power to definitively evaluate the small (4-8%) treatment effect.

9. Study (b) (4) 828-G5780 was a placebo-controlled study performed to demonstrate the therapeutic efficacy of AIGIV administered after the onset of clinical signs of anthrax.

    a. This pivotal efficacy study was performed under GLP at (b) (4) under a study protocol dated 01 August 2008. The in-life portion of the study (including initial quarantine) was 07 August 2008-23 January 2009.

        i. This study was audited by BIMO, who found issues that did not impact the data

submitted to the BLA. The inspection was classified as Voluntary Action Indicated.

b. The primary objective of the study was to assess dose related improvement in inhalational anthrax disease survival between AIGIV and placebo control groups.

    i. The primary endpoint per study protocol was survival at 28 days post-challenge.

        1. Animals were monitored for an additional 62 days for any illness.

c. Sixty-eight (68) juvenile, specific pathogen free NHPs (cynomolgus macaques; *Macaca fascicularis*) of Vietnamese origin were obtained from (b) (4) and surgically implanted with vascular access ports (VAP) and telemetry implants. Animals were individually housed and acclimated with restraint jackets designed for infusion studies.

d. Sixty four (64) of the study animals with a minimum weight of 2.1 kg were randomized to 4 test groups (16 animals per group; 8 males and 8 females), then to a challenge day and challenge order.   The animals were anesthetized with Telazol and exposed to a target (planned) dose of 200 LD_{50} inhaled spores ($1.2x10^7$ colony forming units, cfu). The actual dose was 154 +/- 40 LD_{50}, with a mass median aerodynamic diameter of 1.1 μm.

e. The treatment trigger was detection of PA in serum samples at a concentration >1.5 ng/mL and was verified by (b) (4)

    i. Blood was collected at day -7, 24, 30, 36, 42, 48, 54, 60, 66 and 72 hours post challenge and on days 7, 10, 14, 21, and 28 post challenge.

    ii. CRP levels and standard hematology were also determined.

    iii. Clinical signs (including but not limited to anorexia, lethargy, respiratory distress, and seizure) were monitored every 6 hours on days 0-10 post-challenge, and twice daily (during business hours) on days 11-28.

    iv. Body temperature and activity levels were monitored via (b) (4) implant (b) (4)

f. The test article was AIGIV lot 10602912. The control article was normal human immune globulin intravenous, lot 10703403.

    i. Proposed adult human dosing of AIGIV is 420 U (7 vials)

    ii. Dosing of the test article was based on animal body weight, and a volume of IGIV was administered to yield an equivalent protein load to the 30 U/kg test article group. Total protein loads ranged from 0.16 g/kg (7.5 U/kg group) to 0.65 g/kg (30 U/kg dose group and placebo). Dose volumes ranged from 2.74 in the 7.5 U/kg group up to 11 mL/kg in the 30 U/kg dose group and placebo.

    iii. With the exception of the 7.5 U/kg group, test article or control article was administered at a starting rate of 1.5 mL/kg/hr, increasing by 0.5 mL/kg/hr hourly up to 3 mL/kg/hr. The 7.5 U/kg group received test article at a constant flow rate of 1.5 mL/kg/hr.

    iv. The study was not blinded to treatment. This limitation was discussed when the study was submitted to the IND circa 2011 and it was agreed that since the endpoint was survival the difficulties in blinding (due to volume differences between the treatment arms) justified performing the study in this manner. Reference FDA's 26 January 2011 responses to BB-IND 11982/133.

g. Euthanasia criteria were predefined and included any of the following: moribundity, seizure denoting primary CNS disease, respiratory distress, dyspnea, or forced abdominal

respirations, unresponsiveness, recumbence/weakness, loss of more than 20% body weight, body temperature < 95ºF, or total anorexia with duration > 48 hours.

    i. Study animal disposition was as follows:

        1. Group 1 (IGIV placebo): 7 animals found dead, 8 animals euthanized 1 survivor.

        2. Group 2 (7.5 U/kg AIGIV): 6 animals found dead, 6 animals euthanized, 4 survivors.

        3. Group 3 (15 U/kg AIGIV): 5 animals found dead, 4 animals euthanized, 7 survivors.

        4. Group 4 (30 U/kg AIGIV): 2 animals found dead, 3 animals euthanized, 11 survivors.

h. Pathology

    i. Gross necropsy was performed on animals that died on study. Histopathology was performed on mediastinal lymph nodes, spleen, lung, liver, brain/meninges, adrenal glands, kidney, and gross lesions observed at necropsy. The primary goal was to confirm cause of death as anthrax.

i. Results

    i. All NHP administered AIGIV had a statistically significant increase in survival when compared to the IGIV group.

        1. All animals on study developed toxemia (as defined by circulating PA levels > 1.5 ng/mL). The time to onset of toxemia was comparable across the treatment groups (see Figure 2).

        2. The mean serum PA concentration at the time of treatment was 6.4, 5.3, 16.3, and 7.0 ng/mL for groups 1-4, respectively. The difference in PA values between groups was not statistically significant. See Figure 3.

    ii. With respect to the primary endpoint analysis:

        1. Ninety-four percent (15/16) of group 1 (IGIV) treated animals succumbed to anthrax disease with a median time to death of 127.6 hours.

            a. Eleven group 1 (IGIV) were bacteremic and toxemic prior to treatment; none survived (0%, 0/11).

        2. Seventy-three percent (11/16) of group 2 animals (7.5 U/kg AIGIV) died with a median time to death of 132.8 hours.

            a. One animal was excluded from the survival analysis. This animal (28560) excised its telemetry implant and was euthanized for humane reasons. No evidence of anthrax disease was noted by the pathologist.

            b. Eleven group 2 animals (7.5 U/kg AIGIV) were bacteremic and toxemic prior to treatment. Four animals survived (36%, 4/11).

        3. Fifty-six percent (9/16) of group 3 animals (15 U/kg AIGIV) died with a median time to death of 156.7 hours.

            a. Fourteen group 3 animals were bacteremic and toxemic prior to treatment. There were six survivors in this cohort (43%, 6/14).

        4. Twenty-nine percent (4/14) of group 4 animals (30 U/kg AIGIV) died.

Median time to death could not be calculated since >50% of the treated animals survived.

   a. Two animals (28943 and 28584) were excluded from the survival analysis due to receiving AIGIV prior to a positive PA result.
   b. Ten group 4 animals were bacteremic and toxemic prior to treatment. There were seven survivors (70%, 7/10).

5. A comparison of the Kaplan-Meier curves for the intend to treat set (ITT; includes all animals excluding 28560 from Group 2 and animals 28943 and 28584 from Group 4) to the modified intend to treat set (ITT excluding animals that were not bacteremic and toxemic prior to treatment) are presented in Figure 4 and Figure 5, respectively.

6. The package insert will reflect efficacy for animals that were bacteremic and toxemic prior to treatment according to Table 4.

j. Protocol Amendments and Deviations

   i. There was a single amendment to the study protocol describing two changes to the protocol.

   1. The procedure for calculating median challenge time was clarified by specifying that the median challenge time would be calculated from the end time of all animals challenged within a single challenge day.
   2. The second change corrected a cross-reference in the text so that the appropriate table was referenced.

   ii. Study deviations were provided for study 828- G5780. The study was extensively audited and documented. The majority of the deviations were minor and not uncommon for a study of this type (telemetry errors, documentation errors, slightly late or early observations, minor equipment issues, etc.).

   1. The most significant deviation with regard to potential study outcome was DR-6619. Two animals (28943 and 28584) were treated prior to a positive PA result. For both animals the (b) (4) assay was incorrectly interpreted. These animals were excluded from the survival analysis, so the actual impact to the study result is limited to a minor loss of statistical power in group 4.

10. Essential Data Elements of an Animal Model[30]

   a. Note: This draft guidance underwent substantial revision and an updated version was released in May 2014. Since the Agency had been providing advice and guidance on Cangene's animal model program since 2006, and since the actual preBLA meeting (February 2014) was held with Cangene prior to the release of the updated guidance the previous version released in 2009 is cited here.

   b. The same NZW rabbit and cynomolgus macaque inhalational anthrax models were used to approve raxibacumab with an indication of "treatment of inhalational anthrax" in 2012.

   c. Cangene provided a letter of cross-reference to NIAID's DMF (b) (4) for Anthrax Animal Models. BARDA also performed a meta-analysis on individual animal data based on 37 animal studies, which was included in the BLA and was informative for establishing the 'baseline' anthrax disease model.

d. Characteristics of the Chemical, Biological, Radiological, or Nuclear Agent that Influences the Disease or Condition

    i. Challenge Agent

        1. Bacillus anthracis strain Ames was chosen due to its ubiquitous use in animal challenge studies, its high degree of characterization, and its highly pathogenic nature. Challenge models using the Ames strain have been used to evaluate both therapeutics and vaccines against anthrax disease, and the Ames strain was used in the mail-vectored 2001 bioterrorism attack[31].

        2. The specific material used for the majority of challenge studies to support this BLA originated at (b) (4)

            a. The six spore lots (B31, B33, B35, B36, B37, and B42) used for Cangene-sponsored challenge studies at (b) (4) were characterized based on (b) (4)

            b. The single spore lot used in the Cangene sponsored challenge study at (b) (4)   FY09-025) was characterized based on (b) (4)

    ii. Pathogenic Determinants

        1. The pathogenesis of anthrax disease depends on two plasmids encoding several virulence factors: poly-γ-D-glutamic acid capsule (on plasmid pXO1) and the LF, EF, and PA proteins (on plasmid pXO2). Anthrax strains lacking both plasmids are avirulent, and those lacking pXO2 are attenuated. The capsule acts to inhibit phagocytosis of the vegetative bacteria[32] and may enhance the activity of LT[33] . *B. anthracis* Sterne is a pXO1$^+$, pXO2$^-$ strain and is commonly used as a veterinary vaccine[34], while another pXO1$^+$, pXO2$^-$ strain (STI-1) has been used to vaccinate humans[35]. The mechanism of action of the pXO2 derived exotoxins is described in 3.a, above. *B. anthracis* Ames is both pXO1 and pXO2 positive and is fully virulent in several animal species including man.

    iii. Route of exposure

        1. Humans can be exposed to anthrax through a variety of different exposure routes, with the route impacting the pathophysiology of the disease. The

inhalational route of exposure is most pertinent for biowarfare or bioterrorism involving anthrax spores in part due to the high morbidity/mortality associated with inhalational anthrax compared to other forms of the disease. All challenge studies submitted in support of this BLA utilized aerosol exposure to *B. anthracis* Ames spores generated with

(b) (4)

iv. Quantification of exposure

1. Aerosols in the exposure system were sampled with all-glass impingers (AGI) for spore enumeration. Inhaled doses were calculated for each animal using the resulting aerosol concentration and respiratory data collected with plethsmograph units. The mass median aerodynamic diameter of the aerosol particles was determined with an aerodynamic particle spectrometer and was in the 1-2 µm range for all studies.

v. Host Susceptibility and Response to Etiologic Agent

1. Naturally occurring anthrax has been documented in herbaceous species such as cattle, sheep, goats, antelope, and deer, with human cases usually linked to exposure to an infected animal or animal product. Experimental inhalational anthrax has been demonstrated in mice, rats, guinea pigs, rabbits, sheep, and various species of nonhuman primates. Rodent models are of limited utility in testing anthrax therapeutics due to the sensitivity of mice to strains of anthrax lacking capsule (e.g. *B. anthracis* Sterne) that are avirulent in higher mammals, and rats are sensitive to anthrax toxins but resistant to actual infection[36].

a. Rabbits have been utilized as a model of anthrax infection to test the efficacy of anti-anthrax serums since the late 19th century [37]. They are sensitive to anthrax infection via the subcutaneous and aerosol routes, and pathology is similar between the two routes[38]. The mean lethal dose of anthrax spores in New Zealand White rabbits is 1×10^5 spores and mortality exceeds 99% in NZW rabbits exposed to aerosolized anthrax spores at 200 LD_{50}.

b. Nonhuman primates have also been used for many years as an anthrax model. The rhesus macaque (*Macaca mulatta*) is perhaps the most widely used nonhuman primate historically; however due to supply issues an alternative model was sought. The cynomolgus macaque (*Macaca fascicularis*, previously *Macaca irus*) had been used to evaluate anthrax during the U.S. offensive program and to monitor occupational exposure [39]. Inhalational anthrax in the cynomolgus macaque is highly lethal (although not uniformly so), with 95% of exposed animals succumbing to disease at 200 LD50. The median lethal dose in the cynomolgus macaque is ~6×10^4 spores.

vi. Natural History of Disease

1. Time to Onset of Disease/Condition

a. As stated earlier, the incubation period for inhalational anthrax in

humans has a broad range in part due to the retention of spores in the lungs. The time-to-onset in the Sverdlovsk outbreak was 10-45 days with a mode of 9-10 days[22], and for the 2001 mail-vectored anthrax attack it was 4-6 days (median 4 days). Models suggest that the incubation period is impacted by the spore dose.[21]

b. Inhalational anthrax in rabbits has a more rapid onset than humans. At the spore doses used to support therapeutic development (200 LD_{50}), fever and toxemia (as measured by the presence of PA in the circulation) had a mean onset of 29 (95% CI of 28-30 h, n=164) and 27 (95% CI 26-28 h, n=217) hours, respectively. Clinical signs are limited, and lethargy, lack of responsiveness, and respiratory distress occur late in the disease course. Mean time to death was 75 hours (95% CI of 72-78 h, n=251) and overall survival was 0.8% (2/251).

c. Cynomolgus macaques also show few clinical signs until in the fulminant stage of inhalational anthrax. Lethargy, anorexia, and lack of responsiveness were common but interestingly respiratory distress was not. Fever is difficult to ascertain in this model without telemetry and modeling due to the diurnal rhythm in temperature displayed by this species, but in animals monitored on study 711-G005780 the onset of fever was 36 hours (for 3 consecutive readings above baseline; 10/12 animals) or 63 hours (for 6 consecutive readings above baseline; 8/12 animals). Expanding the dataset to the BARDA meta-analysis, the time to onset of fever increased to 47 hours (95% CI of 44-50 h, n=100). Toxemia was noted at 36 hours (95% CI of 35-37 h, n=145) and overlapped with bacteremia (mean 36 h, 95% CI of 34-37 h, n=164). See Table 7. Mean time to death was 92 hours (95% CI of 86-98 h, n=172).

2. Time Course of Progression of Disease/Condition and Manifestations (Signs and Symptoms)

a. Inhalational anthrax follows a biphasic clinical course in humans. The prodromal stage is characterized by non-specific symptoms including fever, malaise, nausea, and vomiting and may last from hours to several days. There may be a brief period of clinical improvement, followed by a rapid deterioration with the development of acute dyspnea, cyanosis, perspiration, pleural effusions, shock, hemorrhagic meningitis, coma, and death. In some cases the fulminant period of disease may last only hours before death. As stated earlier, there is evidence suggesting that there is a link between the inhaled spore dose and the disease course.

b. In contrast, there does not appear to be a link between disease course and challenge dose in rabbits. Given the rapid disease course in rabbits (mean time to death of 75 hours), there is not a biphasic response and the animals exhibit few overt signs of illness beyond

lethargy, anorexia, and weakness shortly before death. Only 2 out of 251 rabbits (0.8%) survived inhalational anthrax disease in placebo or untreated situations.

c. Cynomolgus macaques also do not exhibit a biphasic illness, although they do have a longer disease course compared to the rabbits. Clinical signs included anorexia, lethargy, and a hunched posture or prostration for one to four days, with the animals becoming less responsive to external stimulus prior to rapidly becoming moribund. Eleven out of 172 cynomolgus macaques (6%) survived inhalational anthrax disease after remaining untreated or receiving a placebo. Weight and age had a significant impact on survival, with older and heavier NHPs demonstrating higher survival rates; due to the high correlation between weight and age in these animals it is not possible to determine which covariate impacts survival.

vii. Trigger for intervention

1. Humans may present with anthrax at any stage of the disease, and given the nonspecific symptoms initial diagnosis requires a degree of clinical suspicion. There are no rapid point-of-use diagnostics approved for anthrax, and laboratory tests or a chest x-ray are needed to confirm the presence of disease.

2. Confirming anthrax disease under the controlled conditions of a laboratory study is more straightforward. Model development studies examined multiple parameters in rabbits (Table 5) and nonhuman primates (Table 6). Clinical signs such as an increase in body temperature would be ideal, and a significant increase in body temperature (SIBT) was considered early in the animal model development process. SIBT tracked closely with the presence of PA and bacteremia, but was difficult to accurately measure in the nonhuman primates due to confounding by their circadian rhythm; this was not a limitation in the rabbit model.   Changes in the ratio of neutrophils to lymphocytes in blood can also serve as an indicator of infection, however the Agency determined that this was not adequate since it was not specific to anthrax disease. PCR can detect the presence of anthrax DNA in the circulation at very early timepoints, and the assay has a short turnaround time. A (b) (4)   assay can detect very low levels of PA in the serum and also has a short turnaround time. Bacteremia is the definitive marker of anthrax disease, however obtaining a positive culture takes a significant amount of time and could delay treatment beyond the therapeutic window of the animal models. SIBT, PA detection by (b) (4) and bacteremia were all examined for potential use as treatment triggers, and in discussion with the Agency (reference BB-IND 11982) PA detection was accepted as an appropriate treatment trigger, with the caveat that a positive bacteremia result should be used to verify actual infection.

viii. Characterization of Medical Intervention

1. Product Class
    a. AIGIV is a purified polyclonal preparation of immunoglobulin containing antibodies directed against *Bacillus anthracis*, mainly directed against PA. AIGIV is prepared from the plasma of human donors previously vaccinated with Anthrax Vaccine Adsorbed (Biothrax). The labeled potency is >60 units per vial, with each unit defined by comparison against a reference standard in a toxin neutralization assay (TNA) such that 1 mU/mL is equivalent to 1 µg/mL of anti-PA activity.
2. Mechanism of Action
    a. Cangene has not performed studies to elucidate the *in vitro* activity of AIGIV, however the mechanism of action is likely due to steric interference where immunoglobulin molecules bound to PA interfere with LF and EF binding and thus prevent toxin (LT or ET) formation. *In vivo*, AIGIV has been shown to neutralize pre-formed toxin in rat models.
3. Pharmacokinetics
    a. Briefly, pharmacokinetic studies were performed in humans, rabbits, and cynomolgus macaques. The details of these studies are described in the pharmacokinetic review memo.
4. Synergy or antagonism of medical products likely to be used in combination
    a. Since antibiotics are the first line therapy against anthrax, the potential for added benefit due to AIGIV above and beyond that expected for antibiotics alone was evaluated using levofloxacin in rabbits and ciprofloxacin in NHPs. These studies failed to reach statistical significance for added benefit beyond antibiotics alone, however there was no interference noted between AIGIV and antibiotics.

ix. Design considerations for Animal Efficacy Studies
1. Endpoints
    a. The primary endpoint for both nonhuman primate and rabbit studies was survival at a predetermined time point. Euthanasia criteria were documented in the study protocols to provide a humane and consistent survival endpoint.
2. Timing of intervention
    a. For the pivotal monotherapy studies, animals were dosed with placebo or AIGIV upon detection of serum PA levels reaching or exceeding 1.5 ng/mL. Each animal was evaluated individually and treated separately upon exhibiting the treatment trigger. The added benefit study in rabbits (study (b) (4) 1182-100011472) used a fixed timepoint of 96 hours for AIGIV or placebo administration due to the specific study design. The use of treatment triggers versus fixed time points was discussed with Cangene during the IND phase of

AIGIV product development.

3. Route of Administration

    a. The route of administration of AIGIV in the animal studies is identical to that used in humans, specifically via intravenous infusion.

    b. Dosing Regimen

        i. Both rabbits and NHPs received doses of AIGIV delivered as a single intravenous infusion, and the PK data in infected and uninfected animals was compared to the PK data from AIGIV use in healthy human volunteers. For a description of the dose scaling, please refer to the pharmacokinetic review memo.

11. Conclusion

    a. The nonhuman primate efficacy studies indicate that it is reasonably likely that AIGIV may be expected to provide a clinical benefit in humans with inhalational anthrax.

        i. Even the lowest dose of AIGIV tested in the monotherapy nonhuman primate model provided a statistically significant survival benefit compared to placebo.

        ii. There is a trend toward increasing efficacy with AIGIV dose in the monotherapy nonhuman primate model, although this trend was not statistically significant.

        iii. Interference between antibiotics and AIGIV was not demonstrated.

            1. Efficacy was comparable between antibiotics alone and antibiotics in combination with AIGIV.

            2. Interestingly, when survival is modeled as a function of AIGIV concentration (using all available nonhuman primate data from studies 828 and 987) low doses of AIGIV are less protective in the absence of antibiotics while higher AIGIV doses appear more protective without antibiotics. This is observed regardless of whether AIGIV total exposure (AUC; see Figure 6) or initial concentration (Cmax; see Figure 7) is taken into account. This may be an artifact of the study design used for study 987, where animals are treated late in the disease course (64 hours).

            3. Likewise, if the survival/dose model is expanded to include both nonhuman primates and rabbits (Figure 8), inclusion of the data from the antibiotic interaction studies tends to decrease the survival probability.

Tables and Figures

**Table 1. PK parameters for TNA data following single IV infusion of AIGIV to male and female cynomolgus macaques**

Dose Group	Sex	Animal ID	Actual Dose (mg/kg)	C_{max} (mU /mL)	T_{max} (day)	Elimination Rate Constant (1/day)	Elimination Half-Life (day)	Clearance (mL/day/kg)	Volume of Distribution (mL/kg)	AUC_{last} (day* mU/mL)	AUC_∞ (day*mU /mL)
5.0 U/kg	Male	C25001	4.84	80.7	0.0417	NR[c]	NR[c]	NR[c]	NR[c]	480	NR[c]
		C26688	4.99	78.2	0.500	NR[c]	NR[c]	NR[c]	NR[c]	342	NR[c]
		C26674	5.04	108	0.0417	NR[c]	NR[c]	NR[c]	NR[c]	392	NR[c]
		C25044	5.07	108	0.0417	NR[c]	NR[c]	NR[c]	NR[c]	484	NR[c]
		Mean	4.99	93.7	0.156	NA[d]	NA[d]	NA[d]	NA[d]	425	NA[d]
		SEM	0.05	8.3	0.115	NA[d]	NA[d]	NA[d]	NA[d]	35	NA[d]
	Female	C26697	4.93	131	0.0417	0.0708	9.80	6.55	92.6	611	752
		C26707	5.09	142	0.0417	NR[c]	NR[c]	NR[c]	NR[c]	901	NR[c]
		C26737	5.17	78.6	0.0417	NR[c]	NR[c]	NR[c]	NR[c]	363	NR[c]
		Mean	5.06	117	0.0417	NA[d]	NA[d]	NA[d]	NA[d]	625	NA[d]
		SEM	0.07	20	0.0000	NA[d]	NA[d]	NA[d]	NA[d]	155	NA[d]
	M/F Combined	**Mean**	5.02	104	0.107	NA[d]	NA[d]	NA[d]	NA[d]	510	NA[d]
		SEM	0.04	10	0.065	NA[d]	NA[d]	NA[d]	NA[d]	74	NA[d]
30.0 U/kg	Male	C26651	28.0	474	0.0417	0.0959	7.23	13.2	138	1980	2110
		C26683	30.7	543	0.0417	0.0862	8.05	10.9	127	2620	2800
		C26668	31.0	537	0.0417	0.159	4.36	12.6	79.1	2380	2470
		Mean	29.9	518	0.0417	0.114	6.55	12.2	115	2330	2460
		SEM	1.0	22	0.0000	0.023	1.12	0.7	18	190	200
	Female	C26741	29.0	577	0.0417	0.0884	7.84	9.54	108	2920	3040
		C22030	29.5	576	0.0417	0.111	6.23	12.8	115	2220	2300
		C21358	29.9	512	0.0417	0.0642	10.8	9.67	151	2850	3090
		C26731	30.6	510	0.0417	0.283	2.45	18.9	66.8	1390	1620
		Mean	29.8	544	0.0417	0.137	6.83	12.7	110	2350	2510
		SEM	0.3	19	0.0000	0.050	1.74	2.2	17	360	350
	M/F Combined	**Mean**	29.8	533	0.0417	0.127	6.71	12.5	112	2340	2490
		SEM	0.4	14	0.0000	0.028	1.02	1.2	12	200	200

a. Tabled values are reported to three significant figures.
b. C26700 and C26666 not reported due to incomplete dosing.
c. Value not reported due to failure to meet acceptance criteria.
d. Not applicable.

Table 2. Summary of survival rates and time to death estimates in study FY09-25.

Group	Survival Rate		Median time to Death (hours)	
	Estimate	Exact 95% Confidence Interval	Estimate	95% Confidence Interval
1 (control; n=5)	0	(0, 0.52)	80	(46,131)
2 (Treated at 48 hrs; n=7)	0.71	(0.29, 0.96)	-	(93.5, -)
3 (Treated at 60 hrs; n=8)	0.38	(0.09, 0.76)	99	(72, -)
4 (Treated at 72 hrs; n=10)	0.10	(0.003, 0.45)	93	(62, 135)
5 (Treated at 84 hrs; n=10)	0.10	(0.003, 0.45)	91	(43, 93)

Figure 1. Kaplan-Meier curves representing time-to-death for study FY09-25; no animals were treated with AIGIV for this study.

### Study FY-025 survival distribution
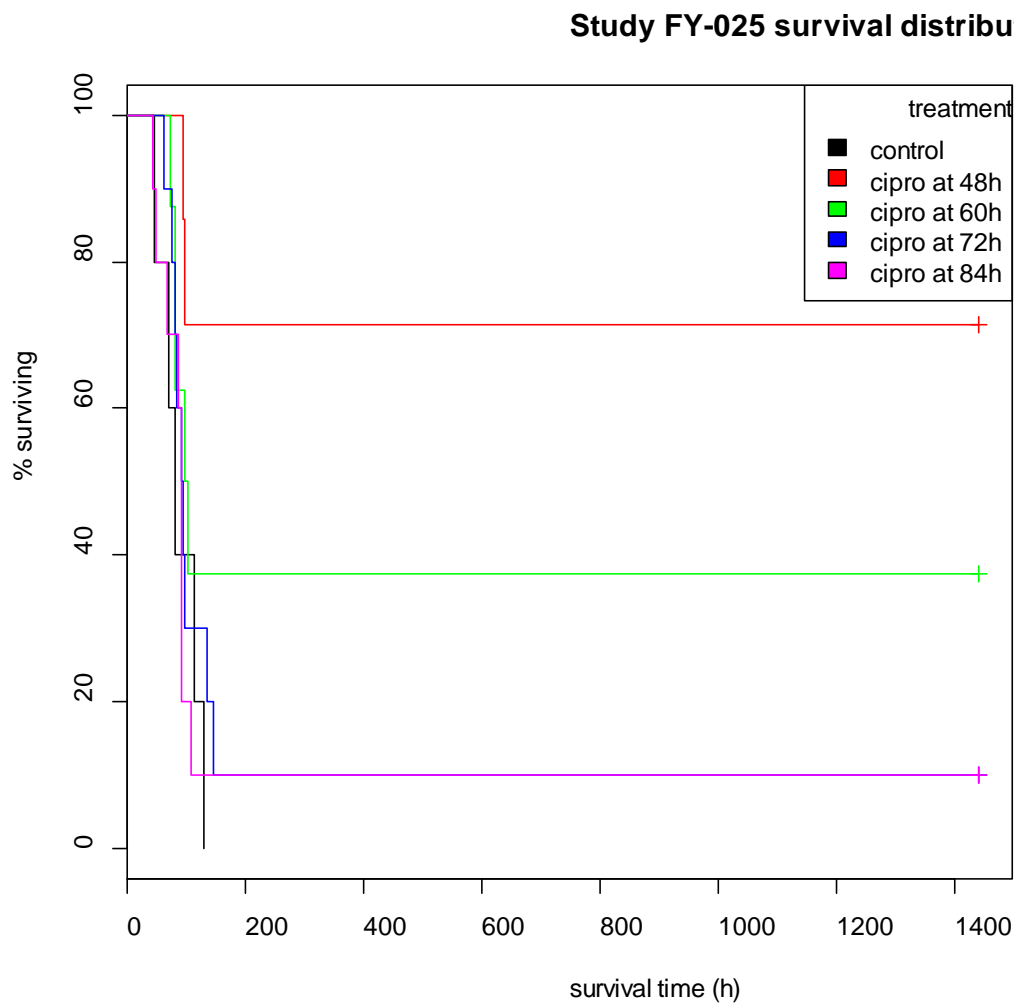
Table 3. Summary of study (b) (4) 987-G005780 survival rates, results of two-sided Fisher's exact test comparisons, and log rank tests comparing Time-To-Death for all treated animals excluding those not bacteremic prior to treatment.
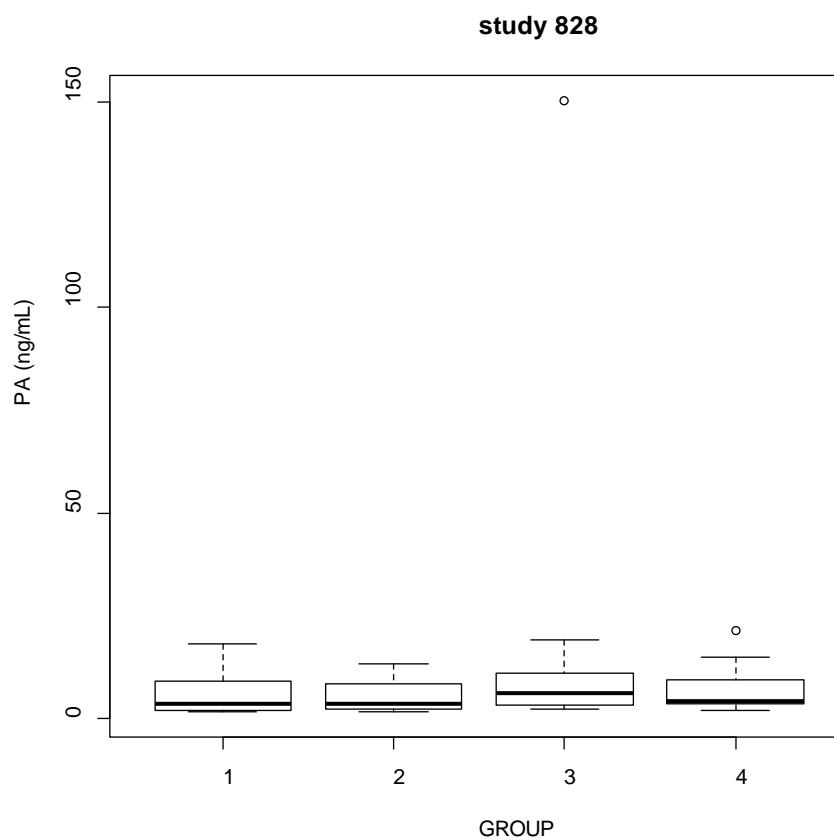
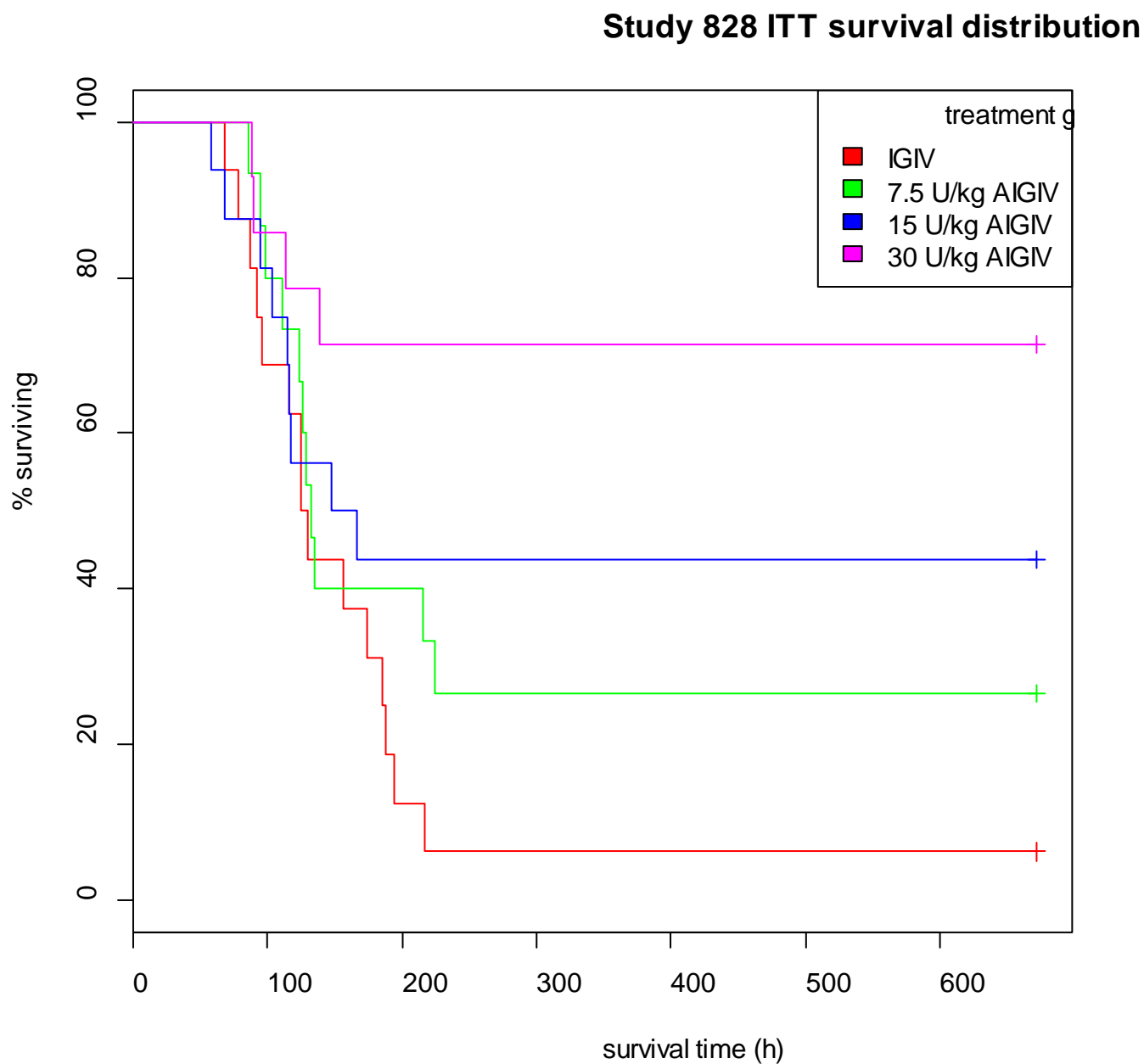| Group	#Survived/ Total	Survival Rate (95% Confidence Intervals)	Two-Sided Fisher's Exact Test P-Values			Log-Rank Test P-Values	
			1	2	3	2	3
1	1/12	0.08 (0.00, 0.38)					
2	9/12	0.75 (0.43, 0.95)	0.0028*				
3	10/12	0.83 (0.52, 0.98)	0.0006*	1.0000		0.6254	
4	11/14	0.79 (0.49, 0.95)	0.0005*	1.0000	1.0000	0.8792	0.7070

*   Significant at the 0.05 level.

Figure 2. Study (b) (4) **828-G005780 time to positive** (b) (4)   **in serum) for each dose group. There was no significant difference between the mean time to positive** (b) (4) **between any groups.**

study 828
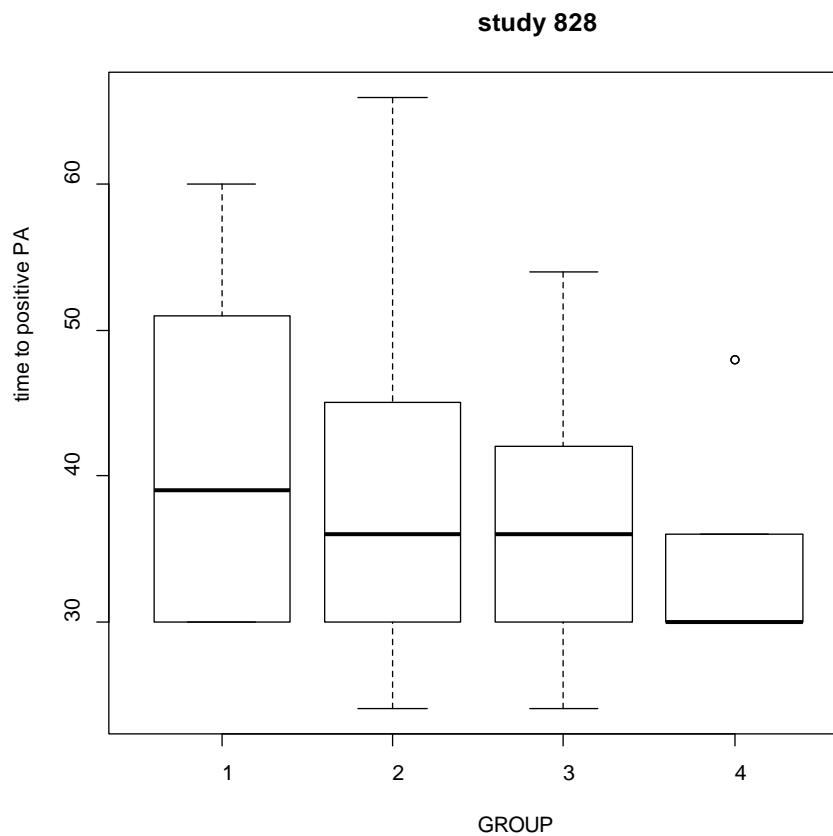
**Figure 3. Study (b) (4) 828-G005780 PA levels at time of treatment for each AIGIV dose group.
There was no significant difference between the mean PA concentration between the groups.**
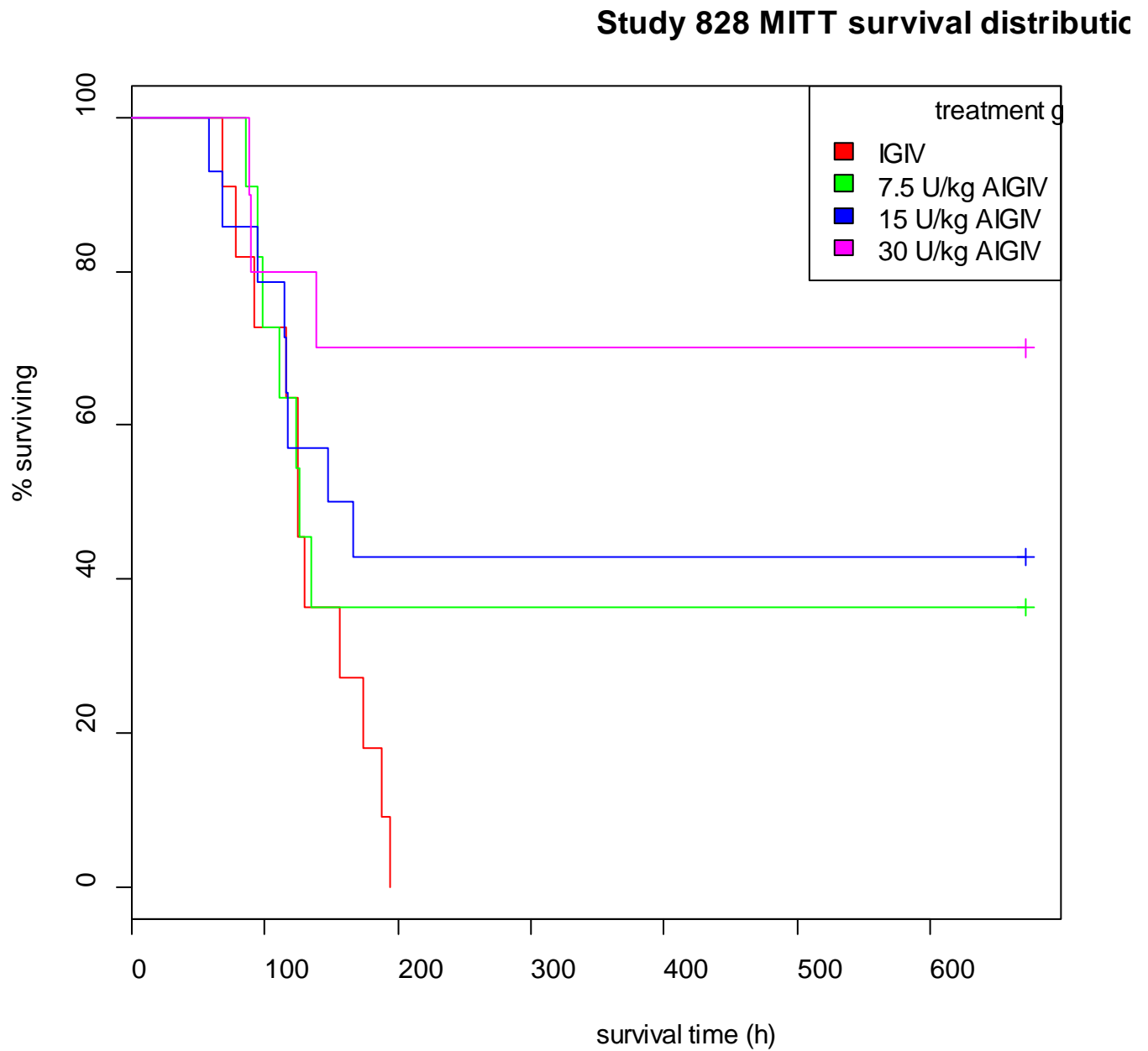
study 828

Figure 4. Kaplan-Meier curves representing time-to-death for the ITT cohort of Study (b) (4) **828-G005780.**

Figure 5. Kaplan-Meier curves representing time-to-death for the MITT cohort of Study (b) (4)
828-G005780.



Study 828 MITT survival distributio

Table 4. Study (b) (4) 828-G005780 summary of survival rates excluding animals not toxemic and bacteremic prior to treatment.

Group	No. Survived/ Total	Survival Rate (95% Confidence Intervals)	One-Sided Fisher's Exact Test P-values					
			Unadjusted			Bonferroni-Holm Adjusted		
			2	3	4	2	3	4
1	0/11	0.00 (0.00, 0.28)	0.0451*	0.0170*	0.0010*	0.0451*	0.0339*	0.0031*
2	4/11	0.36 (0.11, 0.69)		0.5340	0.1349			
3	6/14	0.43 (0.18, 0.71)			0.1846			
4	7/10	0.70 (0.35, 0.93)						

* Significant at the 0.05 level

Figure 6. Survival probability for NHP with inhalational anthrax modeled on AIGIV exposure and presence or absence of antibiotic treatment. The data modeled was obtained from studies 987 and 828.

NHP Survival probability I
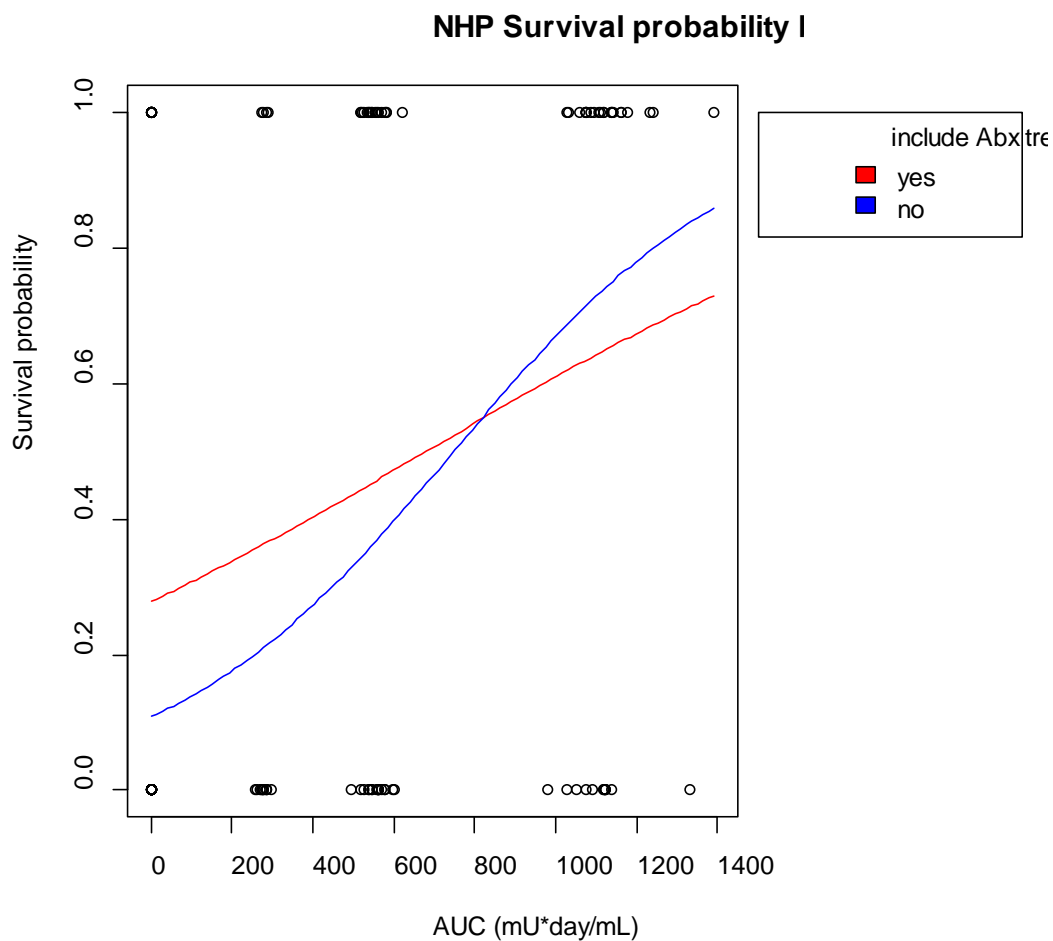
Figure 7. Survival probability for NHP with inhalational anthrax modeled on AIGIV maximum serum concentrations and presence or absence of antibiotic treatment. The data modeled was obtained from studies 987 and 828.
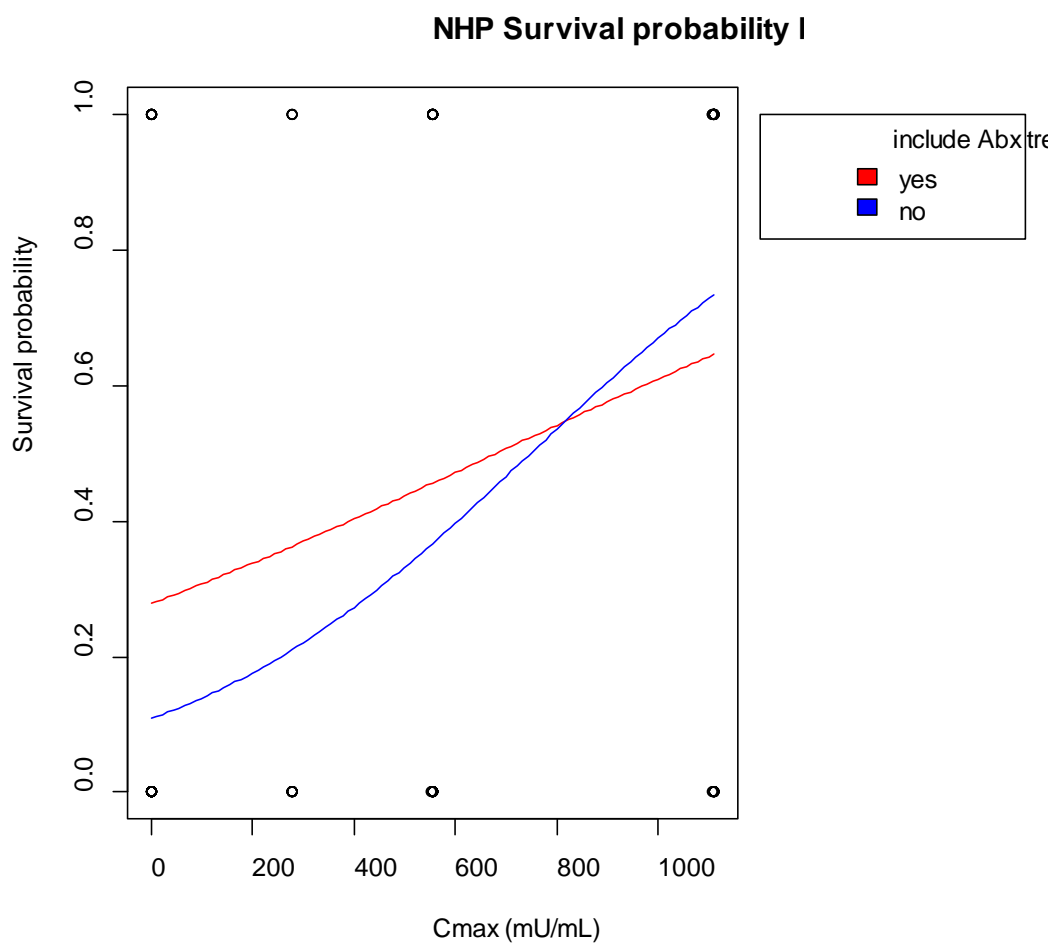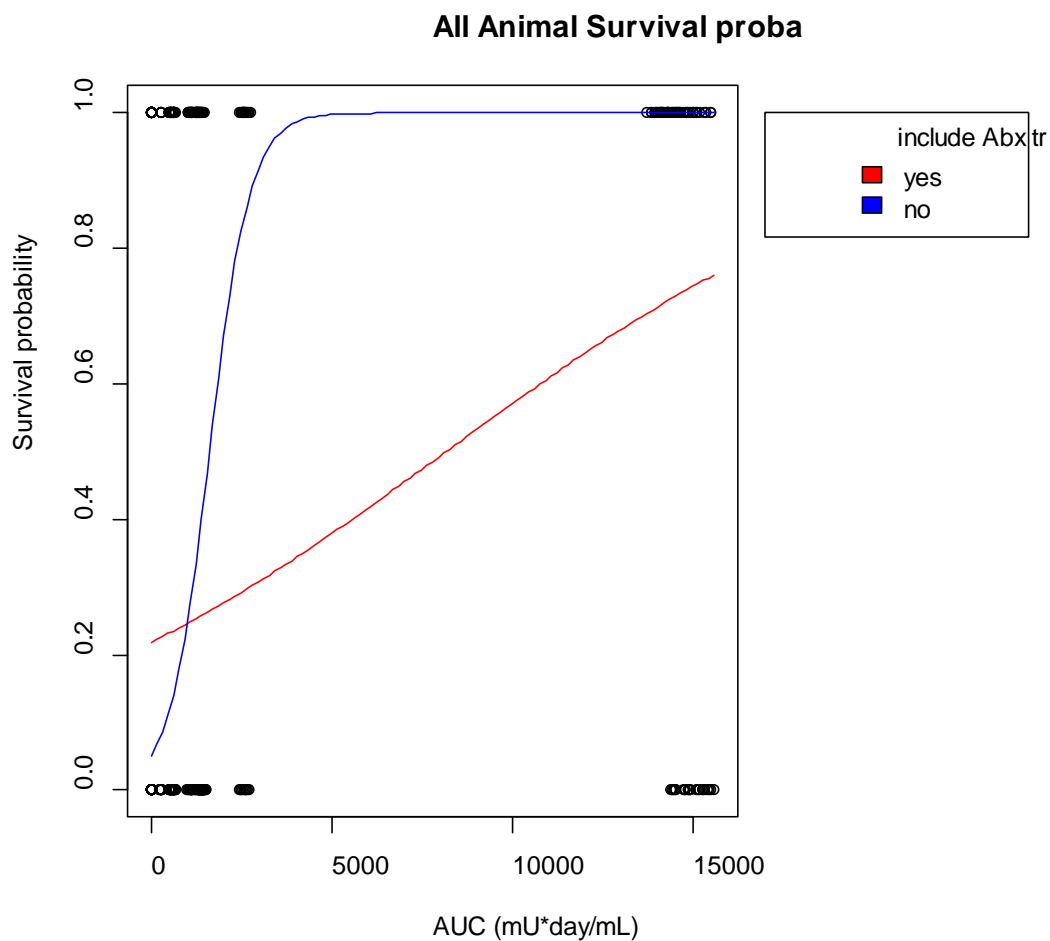
Figure 8. Survival probability for rabbits and NHP with inhalational anthrax modeled on AIGIV exposure and the presence or absence of antibiotic treatment. The data modeled was obtained from studies 677, 828, 898, 987, 1182, and 1207.

All Animal Survival proba

Table 5. Time from anthrax challenge until abnormal observation values in NZW rabbits for parameters analyzed in the BARDA meta-analysis.

Parameter	Number of Animals with Data	Number Abnormal	Number of Studies	Geometric Mean (95% Confidence Interval)	Coefficient of Variation
PA (b) (4)	49	48	8	40.65 (36.25, 45.59)	0.41
PA (b) (4)	217	213	17	26.80 (25.71, 27.94)	0.32
Bacteremia	245	244	21	28.72 (27.38, 30.14)	0.40
SIBT	164	164	17	29.21 (28.12, 30.34)	0.25

Table 6. Time from anthrax challenge until abnormal observation values in nonhuman primates for parameters monitored in study 711-G005780.

Parameter	N	Observations (Incidence)	Mean (h) (95% CI)	Min, Max (h)
SIBT6	12	8[a]	62.70 (51.75, 73.65)	40.97, 82.73
SIBT3	12	10[a]	36.81 (25.56, 48.06)	10.97, 60.28
Activity	12	8[a]	63.90 (30.06, 97.73)	3.97, 127.82
Positive Quantitative Bacteremia	12	12	39.33 (36.47, 42.20)	34.18, 46.57
Positive Qualitative Bacteremia	12	12	41.92 (38.29, 45.55)	34.18, 49.70
PA (Toxemia)	12	12	41.38 (37.67, 45.10)	34.18, 48.90
N/L Ratio	12	12	59.74 (45.18, 74.29)	24.17, 119.13
CRP	12	12	39.99 (29.68, 50.30)	22.53, 72.77
(b) (4)	12	12	48.87 (43.30, 54.45)	36.97, 67.18

[a] 10 of the 12 challenged animals could be included in the temperature and activity analyses.

CRP = C-reactive protein

Table 7. Time from anthrax challenge until abnormal observation values in nonhuman primates for parameters analyzed in the BARDA meta-analysis.

Parameter	Number of Animals with Data	Number Abnormal	Number of Studies	Geometric Mean (95% Confidence Interval)	Coefficient of Variation
PA (b) (4)	90	90	8	32.82 (30.67, 35.11)	0.33
PA (b) (4)	145	144	12	35.97 (34.68, 37.31)	0.22
Bacteremia	172	172	14	35.51 (34.21, 36.85)	0.25
SIBT	100	84	8	47.00 (44.13, 50.16)	0.30

Table 8. Summary of the essential data elements supporting use of the NZW rabbit and cynomolgus macaque inhalational anthrax models to demonstrate efficacy of AIGIV.

Data Element	Animal		Human
	Rabbit	**Cynomolgus Macaque**	
A. Characteristics of the CBRN Agent that Influence the Disease or Condition			
1. Challenge agent	*B. anthracis* (Ames strain)	*B. anthracis* (Ames strain)	*B. anthracis*
2. Pathogenic determinants	Poly-γ-D-glutamic acid capsule mediates invasive stage of infection Anthrax Toxin mediates toxigenic stage of infection	Poly-γ-D-glutamic acid capsule invasive stage of infection Anthrax Toxin mediates toxigenic stage of infection	Poly-γ-D-glutamic acid capsule invasive stage of infection Anthrax Toxin mediates toxigenic stage of infection
3. Route of exposure	Inhalation	Inhalation	Inhalation Cutaneous Gastrointestinal Injectional
4. Quantification of exposure	Bacteremia Toxemia	Bacteremia Toxemia	Bacteremia Toxemia
B. Host Susceptibility and Response to Etiologic Agent	100% susceptible at 200 x LD_{50}	100% susceptible at 200 x LD_{50}	>85% mortality associated with inhalational anthrax without treatment
C. Natural History of Disease: Pathophysiologic Comparability			
1. Time to onset of disease/ condition	1–1.5 days at 200 x LD_{50}	1.5–2 days at 200 x LD_{50}	~4 days
2. Time course of progression of disease/condition	Within 6 days of exposure	Within 10 days of exposure	3.9 days from onset of symptoms
3. Manifestations (Signs and symptoms)	Lethargy Moribundity Respiratory distress Pyrexia	Lethargy Hunched Posture Stool Abnormalities Anorexia	Pyrexia Fatigue Nausea Vomiting Confusion
D. Trigger for Intervention	Detection of protective antigen in the serum by (b) (4) (toxemia)	Detection of protective antigen in the serum by (b) (4) (toxemia)	Suspected or confirmed anthrax infection followed by chest X-ray

Table 8, continued.

Data Element	Animal		Human
	Rabbit	Cynomolgus Macaque	
E. Characterization of the Medical Intervention			
1. Product class	Human plasma derived hyperimmune product		
2. Mechanism of action	Passive immunization		
3. *In vitro* activity	Not applicable		
4. Activity in disease/condition of similar pathophysiology	AIGIV has not been tested in animal models other than inhalation anthrax		AIGIV has been used under CDC expanded access program for treatment of injectional anthrax
5. PK in unaffected animals/humans	$AUC_{0-\infty\ norm}$ (day*mU/kg): 114.0 $C_{max\ norm}$ (mU/mL/U/kg):28.0	$AUC_{0-\infty\ norm}$ (day*mU/mL): 115.9[a] $C_{max\ norm}$ (mU/mL/U/kg):19.0[a]	$AUC_{0-\infty\ norm}$ (day*mU/mL): 423.2[b] $C_{maxnorm}$ (mU/mL/U/kg):27.5[b]
6. PK/PD in affected animals/humans	$AUC_{0-\infty\ norm}$ (day*mU/kg): 46.7 $C_{max\ norm}$ (mU/mL/U/kg):24.2	$AUC_{0-7\ norm}$ (day*mU/mL): 43.5 $C_{max\ norm}$ (mU/mL): 17.8[c]	Not tested
7. PK interactions with medical products likely to be used concomitantly	No interactions when tested in combination with levofloxacin	No interactions when tested in combination with ciprofloxacin	Not tested
8. Synergy or antagonism of medical products likely to be used in combination	Trends towards improved survival	No synergy or antagonism observed	Not tested
F. Design Considerations for Animal Efficacy Studies			
1. Endpoints	**Primary:** Survival **Secondary:** Time to death Incidence and levels of toxemia Resolution of toxemia Resolution of bacteremia	**Primary:** Survival **Secondary:** Incidence and levels of toxemia Resolution of toxemia Resolution of bacteremia	Not applicable
2. Timing of intervention	Detection of PA in serum by (b) (4)	Detection of PA in serum by (b) (4)	AIGIV will be administered to patients with suspected or confirmed inhalational anthrax
3. Route of administration	Intravenous	Intravenous	Intravenous
4. Dosing regimen	15 U AIGIV/kg	15 U AIGIV/kg	420 U AIGIV

Table 8, continued.

Data Element	Animal		Human
	Rabbit	Cynomolgus Macaque	
G. Human Safety Information			
The clinical safety data for AIGIV is primarily from a single clinical trial (AX-001) in healthy volunteers that was designed to assess the safety, tolerability and pharmacokinetics (PK) of three doses of AIGIV. A total of 74 healthy volunteers were administered a single dose of AIGIV intravenously at three dose levels. AIGIV was shown to be safe and well tolerated.			
In addition, a limited amount of safety data was collected from patients treated with AIGIV under a CDC-held Expanded Access Program (EAP, BB-IND 13026). AIGIV was administered to 19 patients for different forms of anthrax; three patients were treated for inhalational anthrax, one patient was treated for gastrointestinal anthrax, and 15 patients were treated for "injectional" anthrax resulting from the use of anthrax contaminated heroin. According to the safety information provided by the CDC to Cangene, infusion of AIGIV was well tolerated with no AEs/ADRs reported post infusion. Six of the 19 patients treated for severe systemic anthrax died (one inhalational and five injectional anthrax patients). Deaths were most likely related to the underlying illness and not to AIGIV administration.			

[a] Both 5 and 30 U/kg doses were used for PK in unaffected cynomolgus macaque calculations

[b] The 210, 420 and 840 U/human doses were used for PK in unaffected human calculations

[c] The 7.5, 15 and 30 U/kg doses were used for the $C_{max\ norm}$ calculation in affected cynomolgus macaque

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