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

To:	To File (BLA STN 125462/0)
From:	Douglas Frazier, Biologist, CBER/DH/LPD/HFM-345
Through:	Dorothy Scott, MD, Chief, CBER/DH/LPD/HFM-345
CC:	Nanette Cagungun, RPM, CBER/DBA/HFM-380
Applicant:	The Cangene Corporation
Products:	Botulism Antitoxin Heptavalent (A, B, C, D, E, F, G) (Equine)
Subject:	Original BLA: Final review

Recommendation

This supplement is recommended for approval, based on review of assay validation and product stability. Final dating and storage conditions will be reflected in the approval letter, and will include lot-specific dating based on currently available potency data, with extensions permitted supported by results of ongoing stability protocols.

Executive Summary

The Cangene Corporation, located in Winnipeg, Manitoba, Canada, has submitted a Biologics License Application for Botulism Antitoxin Heptavalent (A, B, C, D, E, F, G) (Equine), a hyperimmune antibody-fragment product that specifically neutralizes toxins produced by *Clostridium botulinum* and is indicated for the treatment of symptomatic botulism following exposure to botulinum neurotoxin serotypes A, B, C, D, E, F, or G.

Botulism Antitoxin Heptavalent (HBAT) is a "clear-to-slightly-opalescent, colorless-to-pale-yellow" sterile liquid, ----(b)(4)----, containing ---(b)(4)--- of pepsin-digested and purified equine-derived gamma globulin (IgG) polyclonal-antibody fragments ($F(ab)_2$ and Fab) against botulinum neurotoxin serotypes A through G. The HBAT product is formulated in 10% maltose and 0.03% polysorbate 80, contains no preservatives, and is intended for single use by intravenous (IV) administration. Prior to use, the product is to be diluted one in ten (1/10) with 0.9% Sodium Chloride Injection (USP). HBAT is supplied in clear -(b)(4)- glass vials (20 or 50 mL) with ----------(b)(4)---------- rubber stoppers (20 mm), aluminum seals, and plastic flip-top caps. Filling is based on target potency per vial, expressed in units/vial, and based on the amount of toxin-specific neutralizing antibodies to each toxin serotype as determined by the Mouse Neutralization Assay (MNA).

This review determines that:

1. The quality-control assays used for release of BAT lots and to perform stability studies have been successfully validated to have adequate performance, and are are acceptable for their intended purpose;
2. The proposed dating period of ≤ 48 months at -20 ± 5 °C -------(b)(4)------- -20 ± 5 °C followed by 36 months at 2-8 °C is supported by the provided stability data, and may be accepted.

Review

Additional background: Cangene has received FDA Fast Track designation for this product, which is designated internally by Cangene as eBAT NP-018. Cangene has been manufacturing HBAT since 2004 under BB-IND 12052, first at a -(b)(4)- scale and, after an increased requirement for eBAT NP-018 delivery to the Centers for Disease Control and Prevention (CDC) Strategic National Stockpile (SNS), a scale-up to manufacture HBAT at a -(b)(4)- scale. Comparability of the process at both scales was determined under comparability study 740_BAT_07_040.

Botulism Antitoxin Drug Substance is produced from plasma collected from horses immunized with one of the seven botulinum toxin types A through G. Each serotype-specific bulk is processed as a stand-alone manufacturing run, each with its own batch record and separated by line-clearance procedures, yielding a formulated monovalent antitoxin-specific bulk drug substance. There are two sources of equine plasma that have been used for the manufacture of eBAT Drug Substance:

- Phase A Plasma, collected between 1993 and 1996 at BioWhittaker (Walkersville, MD) under contract with the Department of Defense, and
- Phase B Plasma, collected beginning in 2005 from Auburn University (Auburn, Alabama) and Lake Immunogenics (Ontario, New York) under contract with Cangene.

Phase A Plasma was used in the manufacture of -(b)(4)- eBAT Drug Substance batches at the -(b)(4)- scale, while the remaining eBAT Drug Substance batches were manufactured using Phase B Plasma. Auburn University is to remain the sole source for additional Phase B Plasma. All future lots of product are to be manufactured at the commercial -(b)(4)- scale; lots manufactured at the -(b)(4)- scale are currently available in the stockpile.

The manufacturing process for each monovalent eBAT Drug Substance is essentially as follows:

- Plasma pooling
- Solvent/detergent incubation
- Clarification
- Cation-exchange chromatography
- Pepsin digestion
- Anion-exchange chromatography
- Virus nanofiltration
- -(b)(4)- and formulation

There are no reprocessing procedures in the manufacture of eBAT Drug Substance for either the -(b)(4)- or the -(b)(4)- scale. Botulism Antitoxin Drug Substance is routinely stored at ---------(b)(4)---------. The bulk product is dispensed into ------(b)(4)------ are used for stability studies.

HBAT was manufactured under Phase A, at a -(b)(4)- manufacturing scale, and subsequently under Phase B at a -(b)(4)- scale[1]. Cangene performed a comparability study (R%D report 740_BAT_07_040_v2, -(b)(4)- BAT Plasma to -(b)(4)- BAT Plasma Manufacturing Comparability Study, Nov. 2009). A visual assessment of the data finds good comparability between control-test data from heptavalent Drug Product and monovalent Drug Substance; the only difference is the increase in the desired component $F(ab')_2$, the desired form of the active ingredient, i.e., "de-speciated" equine IgG. This change was brought about by an optimization of the pepsin digestion process step, and was assessed by Cangene not only by characterization of the product itself but also by as assessment of clinical safety, which was found to be unchanged. A list of current manufacturing facilities is included (**Appendix 1**).

Assigned review sections include: 1) assay validation including the MNA potency assay, and 2) stability of Drug Substance and Drug Product. Given the long developmental history of this product, and the continued interactions between the Agency and the Sponsor, the potency-assay methodology and performance have been previously assessed and agreed upon; this review serves to confirm that that potency assay performance is consistently maintained. The other assays are mostly using methods previously approved for testing of other Cangene products. Stability is assessed for -(b)(4)- monovalent bulks, frozen heptavalent-antitoxin final product, and final product stored liquid at 2-8 °C.

Potency assay: mouse neutralization assay (MNA)

The Mouse Neutralization Assay (MNA) is an *in vivo* toxin-neutralization assay[2] for quantitation of neutralizing antibodies to *Clostridium botulinum* toxins (serotypes A-G), and is done by the Battelle Medical Research and Evaluation Facility (Battelle), in Columbus, Ohio. The assay was developed by Battelle under contract to the U.S. Army Medical Research and Materiel Command in 1997 from standard *in vivo* toxin-neutralization methodology, i.e., by titering dilutions of antiserum with a fixed quantity of toxin, and using mouse death/survival rates as the endpoint.

----------(b)(4)----------

[1] Cangene states: "In 2004, Cangene Corporation produced -(b)(4)- bulk monovalent serotype runs of BAT at the -(b)(4)- plasma scale, of which -(b)(4)- lot of Heptavalent product and --------(b)(4)------ product were filled (-(b)(4)- Phase A). All these batches were produced using plasma collected at BioWhittaker between 1993 and 1996 (Phase A plasma). Only -(b)(4)-Heptavalent BAT finished product lot was produced due to a shortage of a few of the serotypes. Cangene stored the remainder of the purified BAT bulk serotypes -(b)(4)- and used some for blending with batches prepared at the -(b)(4)- plasma scale in 2006 and 2007 with the newly collected plasma (Phase B) from Auburn University and Lake Immunogenics. -(b)(4)- of Heptavalent BAT was produced from this blending of Phase A and B -(b)(4)- bulks. Starting at the end of 2007, Cangene started producing -(b)(4)- plasma scale monovalent batches using Phase B plasma. The first -(b)(4)- Heptavalent lots at the -(b)(4)- plasma scale referred to in this report were manufactured from the first -(b)(4)- monovalent serotype manufacturing batches. These -(b)(4)- batches were manufactured in a different area than the -(b)(4)- batches in the same facility, using new -(b)(4)- equipment."

[2] Cardella MA. Botulinum Toxoids Botulism: Proceeding of a Symposium. Cincinnati: U.S. Department of Health, Education and Welfare, Public Service, 1964:113-130

----------(b)(4)----------

The source of both botulism toxoid (purified formaldehyde-inactivated *C. botulinum* toxin complexes) and toxins during Phase A plasma production was the ----------(b)(4)----------, with the exception of the Type C toxoid, obtained from ----------(b)(4)----------. The group at the ----------(b)(4)---------- that produced toxoid and toxin for Phase A Plasma is currently known as ----------(b)(4)----------, and is the source of all toxoid and toxin for Phase B Plasma production.

Cangene's three-paragraph description of the origin of the primary reference antitoxins is transcribed in the footnote below, not summarized, because it is densely and concisely written, and cannot usefully be paraphrased[3]. The secondary reference standards used to calibrate the BAT potency assays are these: From ----------(b)(4)----------
---

The US DoD gave a contract to ----------(b)(4)---------- to formulate and calibrate botulinum toxin reference standards for the seven serotypes; the work was completed in 1993-94. -----(b)(4)----- reference antitoxin standards were generated in horses using toxins supplied by ----------(b)(4)----------
---
----------. The reference standards were then calibrated against of purified toxins obtained from ------(b)(4)------ in the mouse neutralization assay (MNA) using a ----------(b)(4)----------
----------. The reference antitoxin standards were then found to have the following potencies:

[3] ----------(b)(4)----------

[--(b)(4)--]

----------(b)(4)----------

[--(b)(4)--]

Reviewer's comments – the values submitted (above) show measurable activity loss over -(b)(4)- by Serotype A but not for the other six serotypes.I It is recommended that a subsequent on-site GMP inspection to Cangene and/or Battelle include a product specialist and specifically go over reference-standard calibration, storage, and dating.

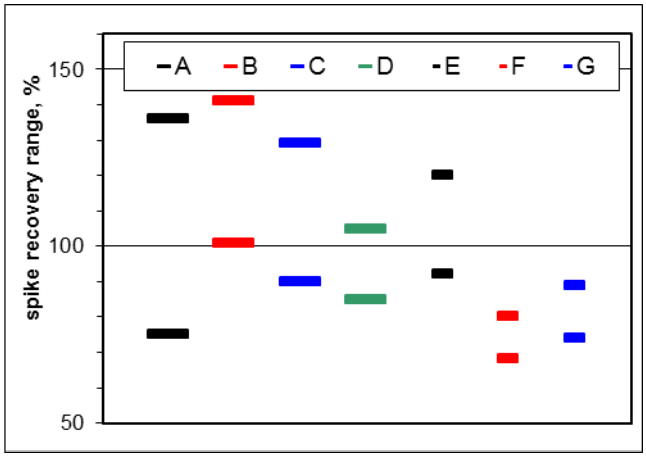
A list of the system-suitability criteria used by Battelle is provided in **Appendix 2**; these are generally suitable for *in vivo* toxin neutralization assay methods.

Cangene provides a potency-method validation report (VAL_MV_101.07_rep_v1, SOP No. MREF.X-014, Validation of the MNA for Heptavalent Botulism Antitoxin Product, dated April 2009), based on separate validation studies for each serotype in monovalent bulk. Validation results for six lots are summarized in the following table:

[--(b)(4)--]

*(b)(4)----------

The above results indicate that the assays for serotypes A, B, and D have significant assay-to-assay variability, while the assay for serotype E has high intra-assay variability; for serotypes C, F, and G, the full amount of imprecision appears to be generated by intra-assay variability, with only slight assay-to-assay variation. Regarding accuracy as % recovery, a plot (below) of the ranges presented by Cangene demonstrates that the assays for serotypes B, C, and E tend to overestimate the amount of neutralizing antibody present, while the methods for serotypes F and G appear to consistently underestimate it. Serotype A can be measured as much as 35% high or 25% low, and has the maximum variability of the seven methods:



Assessment: A potency assay method should be sufficiently accurate and precise as to provide a reliably certain potency value, which allows an appropriate dose of the product to be filled into its final container. This assay methodology, being functional and not an immunoassay, is the most reliable type available, and is done with appropriate controls, sample replicates, and dilutions; its use must be accepted, and its performance limitations must be taken into consideration when calculating target formulation volumes and extrapolating product stability. Cangene uses a -(b)(4)- overfill to make up for both assay variability and potency loss over time.

As a context, the appropriate human dose of HBAT can only be estimated, and depends on the amount of toxin ingested, which cannot even be known without time-consuming tests that, in the case of an emergency situation, cannot be done without further endangering the victim. Therefore, the minimum necessary potency per dose cannot be determined directly; the safest approach is to fill a generous excess, well above what is known to be clinically efficacious. The clinically-effective dose has been estimated from dosages used for the animal test models, since it is unethical to do a prospective clinical trial in humans with a life-threatening toxin. Cangene provides estimates of maximum potential exposure, minimum necessary dose, target overfill, and stability acceptance criteria:

> "The highest serum level of BoNT ever reported in the world was 160 $MIPLD_{50}$/mL (1), this corresponds to a total body BoNT load of approximately 2,400,000 $MIPLD_{50}$. These serum concentrations are an estimate of the maximum amount of BoNT a patient is likely to be exposed to following food-borne intoxication…The human lethal dose of BoNT has not been determined. However, it has been estimated from studies carried out in monkeys (Rhesus macaques and Squirrel) to be approximately 40 $MIPLD_{50}$/kg for BoNT serotype A…Assuming that the average human weighs 70-100 kg, the lethal human dose for botulinum neurotoxin would be approximately 2800-4000 $MIPLD_{50}$. Based on these estimates, the adult dose of eBAT NP-018 (1 vial) contains enough antitoxin to neutralize the estimated human lethal botulinum neurotoxin level thousands of times over at the specification potency as shown in Table 6 [below]… studies have indicated that equine antitoxin to toxin levels is [*sic*] most effective at ratios of at least 30:1…Therefore, the excess eBAT NP-018 relative to the potential toxin levels should maximize BoNT neutralization to prevent further clinical progression, which indicates that the finished product specifications are clinically justified."

Table 6 Capacity of eBAT NP-018 to Neutralize Serum Concentrations of Botulinum Neurotoxins from Various Sources

Serotype	End of Life Specification		Theoretical number of times antitoxin could neutralize BoNT levels from Food-borne Exposure		Theoretical number of times antitoxin could neutralize BoNT levels from Unintentional Botulinum Administration		Theoretical number of times antitoxin could neutralize BoNT levels in estimated Human Lethal Dose	
	Potency (U/vial)	Neutralizing Capacity ($MIPLD_{50}$)	480,000 $MIPLD_{50}$ (US) [5]	2,400,000 $MIPLD_{50}$ (World) [1]	18,000 $MIPLD_{50}$ [4]	36,000 $MIPLD_{50}$ [4]	2800 $MIPLD_{50}$ (Estimated Lethal Dose) [6, 10]	4000 $MIPLD_{50}$ (Estimated Lethal Dose) [6, 10]
A	> 4500	45,000,000	93.8	18.8	2500	1250	16071	11250
B	> 3300	33,000,000	68.8	13.8	1833	917	11786	8250
C	> 3000	30,000,000	62.5	12.5	1667	833	10714	7500
D	> 600	6,000,000	12.5	2.5	333	167	2143	1500
E	> 5100	5,100,000	10.6	2.1	283	142	1821	1275
F	> 3000	30,000,000	62.5	12.5	1667	833	10714	7500
G	> 600	6,000,000	12.5	2.5	333	167	2143	1500

eBAT NP-018 = Botulism Antitoxin Heptavalent (A, B, C, D, E, F, G) – (Equine); $MIPLD_{50}$ = Mouse Intraperitoneal Lethal Dose 50; U = Unit; BoNT = Botulinum Neurotoxin; US = United States

This assay method is the most reliable one currently available, and is done with appropriate controls and sample numbers and dilutions; its performance limitations must be taken into consideration when calculating target formulation volumes and extrapolating product stability. A combined assessment of assay variability, potency loss rates, minimum potency specifications, and target overfills was done by Cangene, which chose a target overfill of -(b)(4)- in the final heptavalent BAT product to compensate for variability and activity loss over time.

Cangene also provides a potency-method validation report (VAL_MV_101.07_rep_v1, SOP No. MREF.X-014, Validation of the MNA for Heptavalent Botulism Antitoxin Product, dated April 2009), based on the following separate validation studies, one for each serotype:

Serotype	Cangene study no.	MREF Study no.	Battelle SOP no.
A	101.01	474-G004744	MREF.X-129.RR
B	101.02	475-G004744	MREF.X-130.RR
C	101.03	476-G004744	MREF.X-131.RR
D	101.04	477-G004744	MREF.X-132.RR
E	101.05	478-G004744	MREF.X-133.RR
F	101.00	414-G004744	MREF.X-116
G	101.06	479-G004744	MREF.X-134.RR

Validation results are summarized below, by lot and by serotype, and include assessments of Accuracy (as recovery of target monovalent bulk potency in formulated heptavalent final product), and Precision (as intermediate precision across all serotypes):

One (1) Page Determined to be Non-Releasable: (b)(4)

----------(b)(4)----------

[--(b)(4)--] [--(b)(4)--]

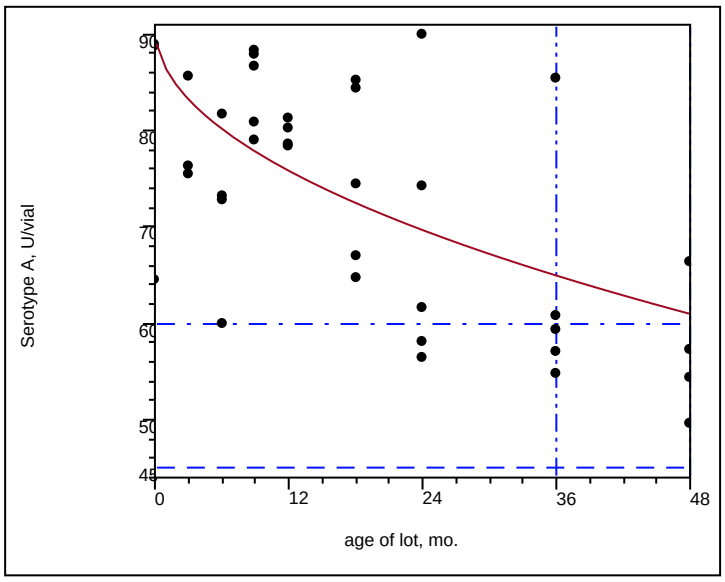
----------(b)(4)----------

[--(b)(4)--] [--(b)(4)--] [--(b)(4)--]

----------(b)(4)----------

[--(b)(4)--]

Assessment: potency recovery for each serotype is acceptable; the assay is sufficiently well validated. It is noted that Cangene proposes a shelf-life at 2-8 °C of 36 months; for Serotype A, which shows the greatest potency loss rate at 2-8 °C, the available stability data indicate a potency loss rate that is nonlinear, and for which the best-fit regression line has a lower 95% confidence limit at 36 months of about 6000 U/vial, or 500 units/vial higher than the minimum required amount:



A = 9070 - 428.8*Sqrt(age, mo.)

Additionally it may be noted that the slope of the trendline for final-product BAT stored at 5 ± 3°C (i.e., 430*√(age, mo.)), is about a third higher than BAT stored at -20 ± 5°C (i.e., 320*√(age, mo.)

Reviewer's comments – the overfill used to make the existing BAT final-product lots is -(b)(4)-; the measured average potency loss for liquid product at 2-8 C is 28-33%. However, no objection is made to this apparent slight discrepancy, due to: 1) the actual greater potency overfill that was generally used for BAT lots, 2) and 2) the ongoing stability monitoring that Cangene has committed to for the manufactured lots.
The MNA's performance is typical of an *in vivo* toxin-neutralization assay method; the high variability is generally confirmed, but has been taken into consideration to determine overfill needed to maintain shelf-life at 2-8 C. The proposed overfill is acceptable.

Other quality control assays
A series of additional assays are used to assess the quality of HBAT monovalent bulk Drug Substance and heptavalent Drug Product, some of which have been previously validated for use with other Cangene products. It may be noted that the earlier HBAT lots of -(b)(4)- batch size had several slight differences in specifications, including: 1) Lower $F(ab')_2$ content ranges, serotype potency limits, and pepsin limit; 2) no heavy metal limits; and 3) a less-detailed visual appearance description. Those specifications hold for the -(b)(4)- lots alone, while the -(b)(4)- lot specifications below are to apply to future production lots. Each assay's validation results are summarized in **Appendix 3**; all results are found to be adequate to support these methods for the stated purposes of release and stability testing.

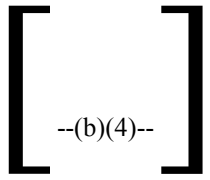
Stability, Bulk Drug Substance (monovalent) (---(b)(4)---**)**

Cangene gives the following description of its stability validation efforts to date:

----------(b)(4)----------

----------(b)(4)----------

----------(b)(4)----------



--------(b)(4)----------

----------(b)(4)----------

Stability studies were conducted on HBAT monovalent Drug Substance batches produced from plasma collected by BioWhittaker Inc. (Walkersville, MD) for the DoD between 1993 and 1996 (referred to Phase A Plasma) as well as on of batches HBAT Drug Substance that were manufactured from plasma collected from a Cangene-managed plasma collection program at Lake Immunogenics (Ontario, New York) and Auburn University (Auburn, Alabama. ----------(b)(4)----------
----

----------:

One (1) Page Determined to be Non-Releasable: (b)(4)

[--(b)(4)--] [--(b)(4)--]

Stability, Drug Product (heptavalent HBAT), -20 ± 5 ºC

Cangene has done a real-time study, i.e., at -20 ± 5 ºC, on three lots produced at the -(b)(4)- scale (Lot Nos. 10804879, 10804882 and 10804483), with 48 months' data generated to date, and on two lots produced at the -(b)(4)- scale (Lot Nos. 2060401 and 10703057), with 86 and 60 months' data, respectively, generated to date. The quality parameters that were monitored included: visual appearance, identity by ---(b)(4)---, molecular size distribution by --(b)(4)--, potency, total protein, -(b)(4)-, and bacterial endotoxins, and sterility. The data were inspected; no or minimal trends were present for all parameters.

The seven botulinum antitoxin serotypes were analyzed via SAS JMP software (**Appendix 5**), and demonstrate that the lots studied show good stability for the time period and storage conditions studied, though somewhat less so for antitoxin against serotype A, the data for which show a clear, apparent first-order or mixed-order degradation-reaction curve.

Regarding the length of the stability studies, Cangene states: "All lots will continue to be monitored on stability at the real time condition of -20 ± 5°C up to the end of the scheduled time point of 120 months." Presumably these results will be used to support future extensions of dating.

Stability, Drug Product (heptavalent HBAT), 2-8 ºC

Stability studies done or underway for monovalent bulk BATs are as follows:

Heptavalent final-product BAT: stability			
Batch size, L	Phase of plasma	Temp.	No. of lots
-(b)(4)-	B	-20 ± 5 °C	26
-(b)(4)-	B	2-8 °C	26
-(b)(4)-	A	-20 ± 5 °C	1
-(b)(4)-	A	2-8 °C	2
-(b)(4)-	A/B	-20 ± 5 °C	1
-(b)(4)-	A/B	2-8 °C	1

Five batches of HBAT (the same ones used in the study above) were stored at 2-8 ºC for ---(b)(4)--- (study no. 250041-04), and tested for the same stability-indicating characteristics. The only appreciable changes in product quality parameters were in the anti-serotype A titer, and to a lesser extent, serotype C

(**Appendix 5**). From these data, Cangene determines that a shelf-life of 36 months at 2-8 ºC is acceptable.

Assessment: the limiting factors in HBAT's shelf-life are: 1) the potency of the final product at release, 2) the expected rate of potency loss under worst-case conditions, and 3) the minimum acceptable potency. Given that frozen product shows little potency loss over time, the most critical parameter is the length of time the product may be stored thawed and refrigerated. So far, the product is shown to be acceptably stable, and should be amenable to future extensions of dating. However, the serotype specificity that diminishes the most rapidly is that of Serotype A; future extensions of dating must be supported by real-time stability data, and no extensions more than a year at one time seem advisable unless more frequent testing is performed in the course of the extended time period.

Dating Period

Cangene states: "The recommended storage temperature of eBAT NP-018 for long term storage is -20 ± 5°C. Botulism Antitoxin Heptavalent may also be held at the storage temperature of 2-8 °C for a maximum of 36 months anytime during the expiration period provided the maximum shelf life of the product is not exceeded. Stability-indicating parameters have all remained within requirements to date and...the potency results for each of the seven serotypes present in heptavalent eBAT NP-018 support an initial shelf life proposal of at least 48 months at -20 ± 5 °C and 36 months at 2-8 °C."

Reviewer's comments: the proposed storage conditions and dating period are:

≤48 months at -20 ± 5 °C *OR*
≤12 months at -20 ± 5 °C followed by 36 months at 2-8 °C.

These storage conditions are supported by the provided stability data, and may be accepted. However, since many lots were manufactured more than 48 months previously, we suggest that Cangene propose a longer dating period, providing that these lots maintain potency and quality parameters. The request for ≤12 months at -20 ± 5 °C followed by 36 months at 2-8 °C is not supported precisely by data, since the 2-8 degree study was performed starting at time zero, not after 12 months of storage at -20 degrees. It is more prudent to allow thawed product for a shorter period of time (24 months) until supportive data is available for prior freezing before storage at 2-8 degrees.

The stability of individual lots can be assessed independently if necessary, rather than setting a universal dating period, as below; if so, the individual lots appear to be sufficiently stable to apply the approach suggested above.

[--(b)(4)--]
(b)(4)----------

[--(b)(4)--]
(b)(4)----------

One (1) Page Determined to be Non-Releasable: (b)(4)

Appendix 1: list of production facilities and addresses for HBAT

Table 1 Name, Address and Responsibility of each Manufacturer

Manufacturer	Responsibility	Registration Number
BioWhittaker Inc. Large Animal Facility [a] 8830 Biggs Ford Road P.O. Box 122 Walkersville, Maryland United States 21793-0127	Phase A Equine plasma collection (between 1993 and 1996)	N.A.
Lake Immunogenics, Inc. [b] 348 Berg Road Ontario, New York United States 14519	Phase B Equine plasma collection (between 2005 and 2011)	N.A.
Auburn University College of Veterinary Medicine Auburn, Alabama United States 36849	Phase B Equine plasma collection (beginning 2005 to current)	N.A.
Cangene Corporation 155 Innovation Drive Winnipeg, Manitoba Canada R3T 5Y3	Manufacturing and in-process testing except for testing contracted to other organizations (see below)	Drug Firm Annual Registration: FEI 3003153579
(b)(4)	Virus validation 9CFR adventitious agent testing of plasma pools	Drug Firm Annual Registration: (b)(4)
Battelle – Medical Research and Evaluation Facility 1425 S.R. 142, JS-3 West Jefferson, Ohio United States 43162	In-process testing by mouse neutralization assay	N.A.
(b)(4)	WNV mini pool testing	FDA Blood Establishment Registration: (b)(4)

CFR = Code of Federal Regulations; FEI = Facility Establishment Identifier; N.A. = Not Applicable; WNV = West Nile Virus

[a] Since 1996, BioWhittaker Inc. is no longer on the supplier list for equine plasma.

[b] Since August 2011, Lake Immunogenics Inc. is no longer on the supplier list for equine plasma.

Appendix 2: System suitability criteria for the mouse neutralization assay (MNA)
(For the determination of botulinum antitoxin titers, as performed by Battelle for the Cangene Corporation)

---(b)(4)---

---(b)(4)---

Eleven (11) Pages Determined to be Non-Releasable: (b)(4)