Memorandum

Date: 14 April 2009

To: File for STN 125335/0

From: Robert Fisher, Staff Fellow, CBER/OBRR/DH/LPD, HFM-345

Through: Michael Kennedy, Team Leader, CBER/OBRR/DH/LPD, HFM-345

Subject: CMC midcycle review, STN 125335/0, Instituto Bioclon *Centruoides* (Scorpion) Immune $F(ab)_2$ Intravenous (Equine)

---

1. Executive summary
    a. Review of STN 125335/0 is ongoing. Deficiencies have been tentatively identified in the CMC section of the submission. These include lack of any process validation, lack of hold time validations, lack of in-process controls, inadequate testing of source equine plasma, and inadequate animal husbandry procedures.
2. Recommended action:
    a. Communicate the following letter ready comments to the sponsor:
        i. NOTE: Due to the length of the information request, it has been amended to the end of this memorandum (see 3.d below)
3. Summary:
    a. STN 125335/0 is an original BLA submission for *Centruoides* (Scorpion) Immune $F(ab)_2$ Intravenous (Equine)
        i. The final drug product is a lyophilized $F(ab)_2$ produced from the plasma of horses immunized with venom extracted from 4 species of North American scorpions: *Centruoides limpidus limpidus, Centruoides noxius noxius, Centruoides lipidus tecomanus,* and *Centruoides suffusus suffusus*.
        ii. Phase II and III studies were performed under IND 10371
    b. I was responsible for reviewing Chemistry, Manufacturing, and Controls section of this submission (Volumes 1.2 to 1.4), with the exception of viral clearance validation, raw materials, stability, specifications, and assay validation.
        i. Sections reviewed include:
            1. Venom Production
                a. Scorpion glands are purchased from --(b)(4)-- suppliers
                    i. Glands are identified and characterized by appearance and the geographical location of where the scorpion was collected.
                    ii. ----------(b)(4)----------
                b. The venom solution is tested against specified release criteria set for ----------(b)(4)----------.
                c. No specific concerns have been noted with regard to venom

production. Stability and batch-to-batch consistency of produced venom will be reviewed on inspection.

2. Plasma Collection
    a. Animal husbandry issues are being reviewed by Dr. Joel Beren, D.V.M. The immunization and bleeding procedure was also discussed with Dr. Beren.
    b. Animals are housed at a Bioclon facility in ---(b)(4)----------.
        i. Details of horse acquisition are not adequate.
            1. It is unclear what level of turnover is present in the Bioclon herd.
            2. Horses that are at least ---(b)(4)--- at the withers, weigh at least --(b)(4)--, have a good physical appearance, and are free of infectious disease may be added to the donor herd. The only infectious agent tested for is -------(b)(4)----------; it is not clear whether any other tests (-------(b)(4)----------) are performed.
            3. It is unclear whether any considerations are given to the geographical location of candidate horses; this may impact what adventitious agent testing and/or vaccinations are relevant.
        ii. Pest/weed control procedures for the animal facility are not described.
        iii. Water/feed monitoring is not described.
        iv. It is unclear whether feed suppliers are audited.
        v. The monitoring of the horse's health status is not detailed. It is unclear whether weight, temperature, incidence of colic, food intake, and other parameters are monitored and recorded.
        vi. ----------(b)(4)----------.
        vii. Deferment periods after veterinary treatment (antihelmintics, antibiotics, analgesics, etc) and prior to blood collection are not described.
    c. The released venom solution is used to immunize horses --(b)(4)----.
        i. The primary immunization series is ----------(b)(4)----------.

1. Additional details on the immunization procedure (volumes, concentration of ---------(b)(4)---, how horses are monitored for adverse events, and the incidence of abscesses at the injection sites) are needed.

ii. Plasma antivenom titers are determined by the method specified in SOP CB-009-01; horses with a minimum antibody titer of ----------(b)(4)---------- are entered into plasma collection.

d. ----------(b)(4)----------.

i. ----------(b)(4)----------

ii. It is unclear whether the equipment used for bleeding horses is sterile and/or disposable.

iii. The description of the bleeding procedure itself is vague. No information is provide on how the skin is prepared, or how a tourniquet is used given the neck bleed site.

iv. The concentration and amount of ---(b)(4)----------bleed is not detailed.

v. The bleeding volumes appear excessive by U.S. laboratory standards. For a --(b)(4)-- horse, Bioclon collects up to ----------(b)(4)---------- period. Although recovered RBCs are administered back to the horses, is not clear how Bioclon monitors these animals for anemia or other health issues.

vi. The batch record indicates that the blood ----------(b)(4)----------

e. RBCs are ----------(b)(4)----------

i. The process of returning RBCs to the horses is not detailed. No information is provided on the viability of the sedimented RBCs.

ii. A hold time for the recovered plasma is not provided.

f. Plasma is tested and released against specifications for ----------(b)(4)----------.

i. Viral testing is not performed.

2 pages redacted: (b)(4)

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

---

c. Process Controls
    i. No process validation was performed, as indicated in STN 125335/10.
    ii. The following are listed as process controls:
        1. Venom release specifications
        2. Horse plasma ---(b)(4)---
        3. Drug substance specifications
        4. Drug product specifications.
    iii. There were no in-process parameters specified for endotoxin, bioburden, or protein concentration.
d. The following information request was submitted to Bioclon prior to midcycle:

1. ---

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

---

2. Please detail the equipment used for blood collection e.g. tubing is it sterile/disposable?

3. ---
---(b)(4)---

---

4. How is the animals skin prepped for blood collection? It should be clipped with a # 40 surgical prep blade, surgical scrub 3 X (Nolvasan or Betadine with alcohol rinses),

Betadine solution applied prior to venipuncture with a sterile disposable blood collection needle of appropriate gauge (16 or larger hypodermic needle e.g. 14 gauge). How is the ----------(b)(4)----------

5. ----------(b)(4)----------

    a. ----------(b)(4)----------
    b. ----------(b)(4)----------
    c. ----------(b)(4)----------
    d. ----------(b)(4)----------

6. The guidelines for collection of blood in laboratory animals vary depending on the frequency of blood collection. If blood is collected every 30 days then a volume of 12 mls/kg/month is acceptable. If blood is collected every 7 days then 3mls/kg/week is acceptable. Bioclon reports the horses weigh ----------(b)(4)----------

7. The process of returning RBC's to the horse is not adequately detailed. ----------(b)(4)----------

8. What is the incidence of abscesses at the site of blood collection?

Horse procurement & Husbandry

9. Please describe in detail the process of acquiring horses to be housed at the Ranch.

    a. In addition to the four points in Item 4,D,1(a), what criteria is used to determine if a horse will be added to the group? Besides the test for -(b)(4)- are there any screening blood tests performed e.g. Chem Screen, CBC and serology?

    b. Are there any geographical considerations as to where the horse has historically been housed?

    c. Is there a medical record that accompanies the horse from the previous owner?

10. Please describe the procedures in place at the ranch for insect, vermin and stray animal control.

11. Are any herbicides or pesticides used at the Ranch?

12. Is the water supply monitored for contamination e.g. herbicides, pesticides, bacterial contamination and heavy metals? If so how frequently is the water monitored?

13. Is the horses' feed (all forms) hay, sweet feed etc monitored for contamination? If so how frequently?

14. Are the feed suppliers monitored? If so do you perform on site inspections of the suppliers?

15. Do you monitor the horse's body weight? If so please describe how the body weight is monitored and how frequently.

16. What is the incidence of colic in the production animals? Is there a correlation between immunization and colic?

17. Does immunization affect the horses feed intake? If so please describe in detail the affect and duration of the affect.

18. Do you monitor the horse's body temperature? If so please describe the technique and frequency. Is there a relationship between elevated body temperature and immunization?

19. Do any of the paddocks or pastures have areas of standing water? Please describe the drainage patterns of the pastures/paddocks and provide elevation diagrams of all areas where the horses have access.

Horse prophylactic immunization.

20. Please vaccinate and appropriately booster all horses and other animals on the farm (cats, dogs) with the appropriate rabies vaccine.

21. Please vaccinate and appropriately booster all horses with the trivalent vaccine covering EEE, WEE and VEE.

QC on production serum:

22. What considerations are given to withdrawal times in respect to agents administered to the horse e.g. anthelmintics, antibiotics, analgesics, expectorants, etc. prior to blood collection?

Venom immunization procedure:

23. ----------(b)(4)----------(b)(4)----------

24. Please clarify the use of your term "challenges". Do you use the word "challenges" to mean ----------(b)(4)----------?

25. What is the total volume administered at the time of immunization -------(b)(4)----------?

26. Post- immunization does the horse experience any more then transient discomfort (pain)? If so is this associated with the ---(b)(4)--- and/or the concentration of venom?

27. What is the incidence of abscesses at the site of immunization?

28. Item 4, D, d mentions after the horses are bled ----------(b)(4)----------

CHEMISTRY, MANUFACTURING AND CONTROLS

29. Please provide the assay SOP (SOP M-CB-011), the assay validation protocol, and the assay validation final report, for your --(b)(4)-- based Identity testing.

30. Please provide the assay SOP, assay validation protocol, and the validation final report, for your Sucrose determination test.

31. For all steps during manufacture where intermediates may be stored for any length of time, please indicate the maximum hold time and storage conditions.

32. Are the -(b)(4)- containers used for the collection of horse blood depyrogenated?

33. Are the -(b)(4)- containers used for the collection of horse blood reused? If so, how are they tested for sterility and endotoxin?

34. Your batch record directs the operator to "----------(b)(4)----------." Where are these mixing steps documented? ----------(b)(4)----------?

35. Your batch record for plasma collection and-------(b)(4)----------, code DM-PR-001, pages 7 and 8 of 18 have spaces for recording of "-----(b)(4)------", and "----------(b)(4)----------". Is -(b)(4)- added to the horse plasma before sampling for ----(b)(4)-----?

36. Your test methods for horse plasma include ----------(b)(4)----------(b)(4)----------?

37. How long do you store the horse plasma before use? At what temperature is the plasma stored?

38. What are your critical process parameters? Have you set in-process controls at these steps? Please note that a typical manufacturing procedure would monitor bioburden, endotoxin, and protein concentration at several steps during the process.

39. ----------(b)(4)----------?

40. ----------(b)(4)----------

41. ----------(b)(4)----------

42. Do you perform any assays to characterize your pepsin or pepsin solution?

43. How long can the pepsin solution -----(b)(4)-----?

44. How many lots (sub-batches) of horse plasma can be mixed per lot of bulk drug product?

45. Are lots (sub-batches) of horse plasma tested for adventitious agents prior to use in manufacturing a lot of bulk drug product?

46. How is the product solution mixed in the reactor? Where are the mixing steps documented in the batch record?

47. Have you set a maximum volume of ---(b)(4)---- that can be used to ----------(b)(4)----------?

48. What is the minimum/maximum time allowed for the pepsin digest? Do you take samples to monitor the progress of the digestion?

49. Do you have data to support that if you ----------(b)(4)----------?

50. What mixing speeds are used? How are these documented?

51. Do you monitor the rate of ammonium sulfate addition? --------(b)(4)----------

52. Have you set limits for the amount of time ----------(b)(4)----------

53. When the solution from the first ammonium sulfate precipitation is clarified, you indicate that "in case of plug of the filtration unit, prepare the filtration unit again using new membranes." How often do the filters clog at this step?

54. Have you established limits for processing time ----------(b)(4)----------?

55. Do you have time limits for ----------(b)(4)--------? Is there a maximum hold time for ----------(b)(4)----------? Are samples taken for bioburden, protein concentration, and endotoxin determination?

56. ----------(b)(4)----------?

57. ----------(b)(4)----------

58. What is the maximum amount of time allowed for ----------(b)(4)----------?

59. Please provide details on how the ----------(b)(4)----------.

60. ----------(b)(4)----------?

61. Do you have a maximum filtration time for ----------(b)(4)----------?

62. ----------(b)(4)----------?

63. Have you validated (or are in the process of validating) the maximum number of uses for your --------(b)(4)----------?

64. How many times may the integrity test for the -----(b)(4)----- be repeated?

65. How do you determine the state of ----------(b)(4)----------?

66. How often is the --------(b)(4)----------?

67. What is the maximum amount of time allowed for the ----(b)(4)----?

68. How do you determine -----(b)(4)-----?

69. How long may the --------(b)(4)----------?

70. ----------(b)(4)----------

71. ----------(b)(4)----------?

72. What volume of water is used to dissolve the excipients? Is WFI or reverse osmosis water used?

73. How many batches require ----(b)(4)----? How many batches require ---(b)(4)---?

74. What is the maximum amount of time allowed for the nanofiltration step?

75. Do you monitor flow rate through the nanofilter?

76. What procedures are followed if the nanofilter clogs?

77. How many lots have failed the post-nanofiltration integrity test? Did you investigate why these lots (if any) failed?