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

Pharmacology/Toxicology Secondary Review
Division of Hematology
Office of Blood Research & Review

TO: The file
CC: Basil Golding, M.D., Director, Division of Hematology, Office of Blood Research and Review (OBRR), Center for Biologics Evaluation and Research (CBER)

FROM: Anne M. Pilaro, Ph.D., Supervisory Toxicologist, Pharmacology and Toxicology Branch, Division of Hematology, OBRR, CBER

SUBMISSION #: STN BLA #125426/0, amendment 8 **and** IND #13551, amendment 65
APPLICANT: Inspiration Biopharmaceuticals Inc., Cambridge, MA
PRODUCT: recombinant, human coagulation Factor IX (IB1001; IXINITY™) for the control and prevention of bleeding episodes in patients with hemophilia B, or for the peri-operative management of patients with hemophilia B

SUBMISSION TYPE: original BLA application, amendment #8 and IND amendment #65, responding to FDA information request by teleconference on August 1, 2012
DATE: November 27, 2012

SYNOPSIS:
This memorandum provides documentation of my secondary review of the pharmacology and toxicology review conducted by Dr. M. Keith Wyatt of amendment #8 to STN BLA #125426/000 and amendment #65 to IND #13551, from Inspiration Biopharmaceuticals, Inc. for their recombinant human Factor IX (code name IB1001; tentative tradename, IXINITY™). Of note, I concur with Dr. Wyatt's original recommendation in his mid-cycle memorandum dated September 17, 2012 that the biologics licensing application may not be approved for marketing until additional information is provided to address potential safety risks of antibody development against contaminating host cell proteins from the Chinese hamster ovary cell line used to produce the drug substance. I also concur with Dr. Wyatt's conclusion in his latest review of amendment #8 to the BLA and amendment #65 to IND 13551 that the design of the pharmacokinetics study in rats is appropriate to compare the exposure and kinetics of the IB1001 (b)(4) before and after manufacturing changes designed to remove the contaminating host cell proteins from the product. However, I do *not* concur with Dr. Wyatt's current recommendation

that an animal immunogenicity study be conducted in mice with the IB1001 product following the manufacturing change, because the study design recommended in his review will not result in data that could provide a meaningful assessment of the ability of the process change to reduce or remove the contaminating immunogenic component or components in the final product. Draft language for a letter-ready comment to replace the comment to the Applicant included in Dr. Wyatt's review is provided below, for inclusion in a response to the Applicant.

LETTER READY COMMENT:

The following comment replaces the letter-ready comment to the Applicant contained in Dr. Wyatt's review, and should be conveyed to the Applicant directly as written:

"1. The design of your proposed rat pharmacokinetics study, submitted as Amendment 08 to STN BLA #125426/000 and amendment #59 to IND #13551 is acceptable to evaluate the expected exposure and kinetics of IB1001 both prior to and following the proposed process change (*i.e.*, to incorporate (b)(4) to remove contaminating host cell proteins from the IB1001 drug substance). However, the rat pharmacokinetics study is not designed to confirm whether the immunogenic component or components, *i.e.* the host cell proteins from the Chinese hamster ovary cell line used to produce IB1001 which elicited the antibody responses in patients, have been sufficiently removed. To further evaluate the effectiveness of the proposed manufacturing change, we strongly recommend that you conduct an additional nonclinical study specifically to assess the development of anti-host cell protein antibodies in response to the pre- and post-process change IB1001 drug substance. We recommend that this study be conducted in rabbits rather than in a rodent species to maximize the chance of development and detection of antibodies to the Chinese hamster ovary cell proteins, and that this study include repeat dosing with both the pre- and post-process change IB1001 drug substance, so that a meaningful evaluation of the effects of removal of the host cell proteins may be performed. We also recommend that you submit your nonclinical protocol for our feedback prior to initiating the study."

SECONDARY PHARMACOLOGY/TOXICOLOGY REVIEW:
Inspiration Biopharmaceuticals, Inc. (Inspiration) has submitted an original BLA application #125426, to support approval of their IXINITY™ recombinant, human Factor IX (rFIX; code name IB1001) product. This product was previously developed under IND #13551 held by Inspiration (the Applicant), and clinical trials are still ongoing under the IND. The established pharmacologic class for IXINITY™ is an antihemophilic blood coagulation factor IX (recombinant), and its proposed indications (from the Applicant's draft labeling) are:

- The control and prevention of bleeding episodes in patients with hemophilia B
- The peri-operative management of patients with hemophilia B

On May 24 2012, Inspiration contacted the FDA to report that 18 of 68 patients receiving IB1001 in ongoing clinical trials had developed measurable titers of antibody directed against host cell proteins (HCP) from the Chinese hamster ovary (CHO) cell line used to

produce the drug substance. Following evaluation of data submitted by the Applicant describing the patient histories, clinical safety profiles, and anti-CHO protein antibody titers after IB1001 treatment and the Applicant's preliminary plans to address the issue, FDA placed the IND on clinical hold on June 26, 2012. The review of the BLA application is ongoing.

In response to the FDA's request to provide additional information on how Inspiration will address the issue of the contaminating HCP in their IXINITY™ product, the Applicant submitted an amendment to both the IND (IND #13551/065) and the BLA (STN BLA #125426/000/008) on October 11, 2012 describing their plans to incorporate an additional (b)(4) step in the manufacture of the IB1001 drug substance, to reduce or remove the HCP from the producer cell line and thereby reduce the immunogenic potential of the drug product, and their protocols to assess the biochemical, biophysical and potency comparability of the post-manufacturing process change IB1001 with the IXINITY™ product used in previous clinical trials. In this same submission, the Applicant also provided a draft protocol for a pharmacokinetic (PK) comparability study in rats designed to evaluate the exposure and kinetics profile of the pre-and post-(b)(4) purified IB1001, and requested that the FDA provide feedback on its adequacy to demonstrate comparability to the pre-process change material.

Reviewer comment: Dr. Wyatt's review memorandum only identifies the BLA submission (STN BLA #125426/000/008) as the subject of his review; however, the identical information was submitted to IND #13551 as amendment #065 on October 11, 2012. Therefore, recommendations to the Applicant that apply to both the IND and the BLA amendments will be addressed in my secondary review.

The focus of Dr. Wyatt's review of STN BLA amendment #125426/000/008 was the proposed rat PK comparability protocol, and its adequacy to support the manufacturing change. In his review, Dr. Wyatt provides a brief summary of the proposed rat PK study, describing the study design, numbers of animals to be tested per group, the timing after IB1001 dosing at which blood samples will be obtained for PK evaluation, and the methodology that will be used to measure IB1001 drug substance levels. His conclusions regarding the rat single-dose PK study are that "…the Applicant's study design is adequate to evaluate the PK profiles of the 'polished' IB1001, and selection of lot #GMP 27 as a comparator is considered appropriate…" I concur with Dr. Wyatt's conclusions that the study design and selection of the test articles are appropriate to evaluate the PK comparability of the pre- and post-manufacturing process change IB1001 product.

Additionally, in his conclusion to his review of the submitted rat PK study, Dr. Wyatt notes that the "…proposed PK study does not address the potential of HCP impurities in the 'polished' IXINITY™ product to induce the formation of antibodies to HCP in animals or in patients, and is not adequately designed to establish the safety of the administered product." He also restates in his recommendation that "To assess the safety of residual HCP, FDA recommends that the Applicant conduct a multi-dose, immunogenicity study in wild-type mice administered 'polished' IXINITY™. The results from this study should definitively demonstrate that the amount of residual HCP

in 'polished' IXINITY™ will not induce the formation of anti-HCP antibodies in animals." Dr. Wyatt has included the following letter-ready comment in his review recommending that the Applicant conduct this study, and offering his suggestions for the study design:

> "Your proposed rat pharmacokinetics study, submitted to BLA 125426/0, Sequence 0008[sic] is not adequately designed to address potential safety concerns associated with the host cell protein (HCP) impurities that you previously reported were present in clinical lots of IB 1001. Therefore, in addition to your proposed comparability studies, we recommend that you conduct a multi-dose immunogenicity study to demonstrate that (b)(4) reduces HCP to levels that will not induce the formation of anti-HCP antibodies in animals. To achieve this objective, we recommend that wild-type mice be administered 'polished' IB 1001 at dose amounts ranging from 2-20 mg total protein/animal, at weekly intervals for 8 weeks. Plasma samples from dosed mice should be analyzed for HCP antibodies at baseline, after the fourth dose, and hen at the conclusion of the study. We recommend that you use (b)(4) using an (b)(4) assay, and that (b)(4) , be included as a positive control."

Although Dr. Wyatt's conclusion that the Applicant's proposed rat PK study is not sufficiently designed to address either the safety or the potential immunogenicity of the "polished" IXINITY™ product is correct, the additional nonclinical study to evaluate the immunogenicity of the post-manufacturing change IB1001 product as recommended in his review is not likely to provide useful information that will address his concerns, for the following reasons:

1. The nonclinical studies submitted to the original BLA and the subject of Dr. Wyatt's previous reviews did not evaluate plasma or serum samples from IB1001 dosed mice, rats, dogs, pigs or non-human primates for antibody development to the CHO host cell proteins (for a listing of the studies reviewed, please see Appendix 1 of this review). In addition, no animal studies were conducted with the specific clinical lots of IB1001 that appear to be associated with the anti-HCP antibody development in treated patients. Therefore, there is no baseline from which the immunogenicity of contaminating HCP in the pre-process change material or the recommended dosing schedule can be established, and used to compare to the results obtained with the "polished" IB1001 drug substance.
2. As recommended, Dr. Wyatt's study design does not include a group of animals treated with the pre-manufacturing process change IB1001 product; therefore, the effectiveness of the process change ((b)(4)) in removing the immunogenic component or components in IB1001 cannot be evaluated, since no direct comparison of the immunogenicity pre- and post-process change will be possible.

3. The Applicant may have other assay methodology than the (b)(4) assay recommended by Dr. Wyatt that could provide a more robust evaluation of the immunogenic responses to residual HCP in the pre- and post-process change IB1001 product. Additionally, they may propose different positive control and detection reagents to use in their assay. Therefore, the decision on which assay or assays to use, and which control articles to test should be left to the Applicant.
4. In discussion with the BLA review committee chairman on Nov 20, 2012, Dr. Roman Drews identified the use of wild-type mice to test the immunogenicity of the contaminating HCP in the IB1001 product as problematic. Specifically, Chinese hamsters (from which the CHO producer cell line for IB1001 was derived), mice, and rats are all members of the taxonomic order *Rodentia* and family *Muridae* of mammals. Therefore, his concern is that mice and rats may not be phylogenetically distinct enough to develop a robust immune response to the hamster cell proteins present in IB1001. After discussion with Dr. Drews, the rabbit (order *Lagomorpha*, family *Oryctolagus*) will be recommended as the test animal species in which to evaluate the immunogenicity of the pre- and post-manufacturing process change IB1001 product.

CONCLUSION:
In conclusion, I concur with Dr. Wyatt's decision that the rat PK comparability study is appropriately designed to evaluate the exposure and PK profile of the pre- and post-manufacturing process change IB1001 products. I also concur with his conclusion that the rat PK study does not address either the safety of the post-change IB1001 product, nor confirm whether or not the proposed manufacturing change will reduce or remove the immunogenic CHO host cell protein component or components from the IB1001 drug substance. Although I support Dr. Wyatt's recommendation that an additional animal study should be conducted, I do *not* concur with the design of the study as recommended by Dr. Wyatt, since it is not likely to provide informative data. Therefore, his letter-ready comment will be replaced with a more general recommendation that the sponsor conduct a comparative immunogenicity study of the pre- and post-process change IB1001 product in rabbits. The draft language to be conveyed to the sponsor is included in the letter-ready comment above.

Reviewer comment: It should be noted that the rabbit study design as recommended in this review is not intended to evaluate the comparative immunogenicity of the IB1001 product itself in test animals before and after implementation of the proposed manufacturing change. The proposed rabbit study is also not intended to detect a statistically significant, nor clinically meaningful reduction in antibody responses to the contaminating host cell proteins in the pre- or post-process change IB1001 product. The generation of an antibody response in animals cannot be extrapolated to predict whether or not a similar response will be observed in human subjects treated with the "polished" IB1001. Instead, the proposed rabbit study is intended only to provide a general evaluation and potential confirmation that the process change, specifically the (b)(4) step in the drug substance manufacturing, will reduce or remove the CHO host cell proteins that resulted in the immunogenic response in patients treated with IXINITY™.

Appendix 1. Preclinical studies previously reviewed for this BLA application.

The following preclinical studies were submitted to the original BLA #125426, and were the subject of Dr. Wyatt's earlier, mid-cycle review (dated June 20, 2012):

1. Evaluation of a novel human recombinant Factor IXX preparation in a canine model of hemophilia B. Study #IB1001-PT-019.
2. Thrombogenicity evaluation of IB1001, Benefix and Mononine following a single intravenous administration in (b)(4) rats. Study #IB1001-PT-010.
3. Thrombogenicity evaluation of IB1001 following a single intravenous administration in (b)(4) rats. Study #IB1001-PT-011.
4. Thrombogenicity of Factor IXaβ following a single intravenous administration in (b)(4) rats. Study #IB1001-PT-018.
5. 24-hour evaluation of the plasma pharmacokinetics of six Factor IX samples following a single intravenous dose to (b)(4). Study #IB1001-PT-016.
6. 24-hour evaluation of the plasma pharmacokinetics of six Factor IX samples following a single intravenous dose to (b)(4). Study #IB1001-PT-017.
7. Pharmacokinetic properties of IB1001 drug substance in normal rats: Comparison between "original", "refined", and "commercial" cGMP manufacturing processes. Study #IB1001-PT-R-022.
8. 14-day evaluation of the safety of Inspiration Biopharmaceuticals Factor IX following a single intravenous administration in (b)(4) rats. Study #IB1001-PT-005.
9. 14-day evaluation of the safety of Inspiration IB1001, Benefix and Mononine following a single intravenous administration in (b)(4) rats. Study #IB1001-PT-009.
10. 14-day evaluation of the safety of Inspiration Biopharmaceuticals Factor IX following a single intravenous administration in (b)(4) dogs. Study #IB1001-PT-006.
11. 28-day evaluation of the safety of Inspiration Biopharmaceuticals Factor IX following repeat intravenous administration in (b)(4) rats. Study #IB1001-PT-008.
12. Local tolerance evaluation of Inspiration Biopharmaceuticals Factor IX. Study #IB1001-PT-007.
13. Local tolerance of Inspiration Biopharmaceuticals Factor IX. Study #IB1001-PT-021.

According to the Applicant and based on the information provided in the study reports cited above and reviewed previously by Dr. Wyatt, all *in vivo* pharmacology, pharmacokinetics, single- and repeat-dose toxicology testing of IB1001 met the criteria to demonstrate the nonclinical safety and biologic activity of IXINITY™, and support its intended clinical use.