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

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

To:	File BLA STN 125426 (mid-cycle)
Reviewer:	M. Keith Wyatt, Ph. D., Pharmacologist, CBER\OBRR\DH
Through:	Anne M. Pilaro, Ph. D., Supervisory Toxicologist, CBER\OBRR\DH
Applicant:	Inspiration Biopharmaceuticals
Product:	Ixinity, recombinant coagulation Factor IX
Purpose:	For prophylaxis and on-demand treatment of hemophilia B
Date received:	June 20, 2012

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Recommendation:

Pre-clinical studies submitted to BLA STN 125426 by the Applicant have all been reviewed and are considered adequate by the Pharmacology/Toxicology discipline to support licensure of recombinant Factor IX (Ixinity). However, additional *in vitro* or *in vivo* pre-clinical studies may still be required prior to licensure to demonstrate that recent modifications to the Ixinity (b)(4) method can be incorporated into the manufacturing process, without affecting the toxicity or structural and functional integrity of Ixinity. An additional repeat-dose toxicity, toxicokinetic and immunogenicity study in animals may also be required to demonstrate that host cell protein levels in the final product are sufficiently low to avoid inducing an immune response to these impurities in patients administered Ixinity.

2. Key issue:

In a submission to the BLA on May 30, 2012, the Applicant provided results demonstrating that elevated host cell protein (HCP) levels in the Ixinity drug product correlated with an increased incidence in the formation of anti-HCP antibodies in patients receiving Ixinity treatment. While the increased anti-HCP titers were not associated with any adverse or severe adverse events, the presence of these antibodies was a potential safety concern. To address the elevated HCP levels in the Ixinity product, the Applicant has conducted a root cause analysis and has modified the (b)(4) protocol to include a (b)(4) . The effect these changes may have on the quality and efficacy of Ixinity will be characterized analytically during comparability studies. The Pharmacology/Toxicology discipline will also reserve the right to request additional *in vitro* and *in vivo* studies if deemed necessary, to fully evaluate the safety and efficacy of Ixinity produced using the modified (b) (4) procedure.

3. Comment to be included in the pending CR letter:

1) We advise that additional, pre-clinical pharmacokinetic, efficacy and toxicology studies in wild-type rats and/or HemB KO mice may be necessary to confirm the structural and functional integrity of the post-process change Ixinity product (i.e. (b)(4)). We also advise that an additional, repeat-dose toxicity and toxicokinetics study(ies) in animals may be required. We recommend that these study(ies) evaluate both the pre- and post-process change material in the same study(ies), so that a meaningful evaluation of the effects of removal of the host cell proteins on the safety and potential immunogenicity of Ixinity may be performed.

4. Executive Summary:

Inspiration Biopharmaceutical, Inc. (Applicant) has developed a recombinant human Factor IX drug product (tentative trade name: Ixinity) for use as on-demand or prophylaxis treatment of hemophilia B, at a dose of 75 IU/kg. Prior to the first-in-human trials, the safety of the Ixinity (b)(4) drug product (DP) was assessed during acute and repeat-dose toxicity studies in rats. While results from the acute-dose study suggested Ixinity was safe, unexpected thrombogenicity was observed in rats repeatedly administered Ixinity at the clinically relevant dose of 65 IU/kg. An investigation eventually linked the thrombogenicity to faulty catheters; therefore, in 2008 this effect was not considered drug-related by the former toxicology reviewer. Results from an additional repeat-dose study conducted in rats at a dose of 205 IU/kg, and thrombogenicity studies in rats conducted with Ixinity (b)(4) alone or in combination with activated recombinant Factor IX all confirmed product safety. Of note, Ixinity was only evaluated for potentially acute and sub-chronic toxicity in rats, and in no other animal species.

Results from comparative efficacy studies conducted in hemophilia B dogs administered Ixinity, Mononine® or BeneFIX® demonstrated similar decreases in whole blood cell clotting times and aPTT after treatment with each of the products, at clinically relevant doses. Pharmacokinetic (PK) studies in hemophilia B dogs and rats administered Ixinity DS or Ixinity DP yielded acceptable PK profiles.

Dermal toxicity and local tolerance studies conducted in rabbits administered Ixinity at 75 IU/kg revealed some inflammation and edema at the injection site, but these results were considered acceptable by the reviewer. Overall results from the toxicity studies suggest Ixinity will not present an excessive risk to patients at the clinically relevant dose, but specific, safety pharmacology assessments (*e.g.*, cardiac) as further confirmation were not performed.

Ixinity immunogenicity was assessed by measuring anti-Ixinity antibody levels in rats following repeated administration. As expected for animal studies, detectable levels of anti-human FIX antibodies were observed in several rats; however, these antibodies were not further characterized for inhibitor activity using the (b)(4) assay or other valid detection method. Moreover, Ixinity toxicokinetics were not evaluated to measure the potential decreases in Ixinity exposure that may have suppressed any potential toxicities during these repeated dose studies in rats.

Preclinical studies to measure antibody formation against the host cell protein (HCP) impurities from the Chinese hamster ovary cell line used to express Ixinity were not conducted prior to Phase 1 of the IND. While this omission was not a concern initially, HCP impurity levels in Ixinity were reported to progressively increase in product manufactured and used during the clinical trial. This increase in HCP resulted in an unacceptable frequency of treated patients demonstrating formation of anti-host cell protein antibodies during phase 2 and 3 clinical trials. Although no AE or SAEs were reported, antibody formation was considered a safety risk and a clinical hold was placed on the trial. A root-cause analysis conducted by the Applicant tentatively linked the increased antibody response to a change in the (b)(4) supplier; however, this association has not been confirmed and the ongoing trial remained on clinical hold as of August 27, 2012.

At the time the BLA was filed, the Applicant introduced a concerted effort to reduce the HCP impurity levels, using a (b)(4) . The initial results following (b)(4) showed lower impurity levels, visualized qualitatively by (b)(4) . Although these results appeared promising, additional questions remain about the effect the (b)(4) may have on the efficacy and PK of Ixinity. To address these questions, the Applicant has offered to voluntarily conduct additional animal PK studies to ensure the structural and functional integrity of Ixinity following (b)(4) (b)(4) . Additional studies to improve and scale-up the (b)(4) procedure, as well as to validate the purity of Ixinity using (b)(4) were ongoing as of August 27, 2012.

A summary of pre-clinical results is presented in the table that follows.

5. Summary of pre-clinical toxicology and pharmacology results submitted to BL 125426 to support licensure of Ixinity.

	Study #	Dose	Result (NOAEL)
TOXICOLOGY			
Repeat dose, rats, 8 doses	P0907010, IB1001-PT-008	~65 IU/kg	65 IU/kg
Repeat-dose, rats, 28 doses	IB1001-PT-R-025	~205 IU/kg	205 IU/kg
Acute single dose, rats, 14-day	P0207002, IB1001-PT-005	50, 100 and 200 IU/kg DP	50 IU/kg
Acute single dose, rat, 14 day	IB1001-PT-009 P0108001	348 IU/kg DS and DP	Not claimed
Acute dose, dogs, 14 day	P0207003, IB1001-PT-006	50, 100 and 200	200
Local Tolerance, rabbits Dec 2007	IB1001-PT-007, P0207004	250 IU/kg DS, IV 125 IU/kg DS, IM	100
Local tolerance, rabbits	P0510001, IB1001-PT-R-021	0.4 mg/kg, IV 0.3 ml, PV	??
SAFETY PHARMACOLOGY	Not Performed		
THROMBOGENICITY			
Thrombogenicity & aIX, rats	P081015, IB 1001-PT-018	100, 300 and 1000 Iu/kg	1000 IU
Thrombogenicity, rats	IB 1001-PT-011 P0908016	1000 IU/kg DP	1000 IU
Thrombogenicity, rat	IB1001-PT-010 P0108012	100, 300 and 1000 IU/kg DP or DS	1000 IU
PHARMACOKINETICS			
PK, rat, normal and refined	IB1001-PT-R-022, P090402,	0.4 mg/kg	α phase, 1 hr β phase, 5.4 hr
PK, rat	IB1001-PT-017	~0.4 mg/kg	
PK, rat	IB1001-PT-016 P070705	68 IU/kg DS 81 IU/kg DP	
Toxicokinetics	Not Performed		
PK/PD Crossover study in Canine model of hemophilia	IB 1001-PT-019	50 IU/kg 100 IU/kg	α phase, 3 hr β phase, 15.8 hr
IMMUNOGENICITY			
Immunogenicity (*e.g.*, Inhibitor assay and host cell protein assay	Not Performed		
MUTAGENICITY *	Not Performed		
CARCINOGENICITY*	Not Preformed		
REPRODUCTIVE TOXICOLOGY*	Not Performed		

* not performed based on provision in described in ICH S6, DART, mutagenicity and carcinogenicity studies