

Eradication of factor IX neutralizing and anaphylactic inhibitors in a patient with severe haemophilia B using cyclophosphamide immune suppression and factor IX desensitization

Plasma-derived and recombinant factor IX (FIX) replacement therapy has changed the landscape for treatment of patients with haemophilia B (PWHB). FIX replacement increases haemostasis in PWHB and reduces risk of bleeding-related morbidities such as hemarthroses, intramuscular haematomas and intracranial haemorrhage. Regrettably, 1.5%-3% of PWHB develop neutralizing antibodies (inhibitors) towards FIX concentrates.¹ While most mutations causing haemophilia B are missense point mutations, large and full gene deletion mutations are associated with a higher risk of inhibitor and anaphylaxis formation.² FIX inhibitor formation is a rare but severe complication of haemophilia B that undermines interventions for acute haemorrhage by preventing the replenishment of therapeutic levels of FIX. The development of FIX inhibitors increases patient proneness to bleeding-related morbidities and life-threatening anaphylaxis.¹ Many of the therapeutic options for eradication of inhibitors in PWHB are borrowed from treatment regimens of inhibitors in patients with haemophilia A and utilize composites of immunomodulatory drugs, immunosuppressive drugs and immune tolerance induction.³ Current protocols offer limited success with HB due to risk of anaphylaxis and relative resistance to ITI therapy. Furthermore, high-dose immune tolerance induction (ITI) can result in nephrotic syndrome.⁴ We report our effective management of a child with severe haemophilia B who had FIX neutralizing antibodies and anaphylaxis to FIX concentrates using a desensitization protocol and immune eradication with cyclophosphamide.

An 8-year-old boy with severe haemophilia B secondary to a deletion of the entire F9 gene (HGVS Variant: c.1-?_1386+?del) and inhibitor had been followed by another hospital. According to notes obtained from this institution, this patient was diagnosed with severe haemophilia B at 3 months of age due to easy bruising and was prescribed replacement therapy with BeneFIX (recombinant FIX; Pfizer). His first anaphylactic reaction occurred at 15 months of age when a low-titre inhibitor (1.8 Bethesda Units [BU]) was detected. Prophylactic treatment with NovoSeven® (recombinant activated factor VII; rFVIIa; Novo Nordisk, Inc) was prescribed at that time. His inhibitor decreased to 0.8 BU 4 months following the withdrawal of rFIX (age 19 months), and this patient was negative for neutralizing inhibitor at 3 years and 1 month of age. The patient presented with his first subdural haematoma at age 7 years and 3 months of age. Administration of rFVIIa at this time resulted in radiologic improvement assessed by head CT scan. Approximately 2 months after

his first subdural haematoma (age 7 years and 5 months), this patient presented to the emergency department with a headache and vomiting. Imaging revealed new areas of subdural haematoma. Of note, the patient had not received rFVIIa despite the prescription of prophylactic rFVIIa. At this time, a BeneFIX sensitization was attempted. This patient was challenged with a low dose for 4 days with a titration up to the full dose for 5 days. After 4 days of being on the full dose, the patient experienced anaphylaxis. BeneFIX was discontinued, and full therapy for intracranial haemorrhage with rFVIIa was initiated.

This patient was first seen at our institution 14 months later (age 8 years and 7 months) and was instructed to continue use of rFVIIa. Four months later (age 8 years and 11 months), this patient presented with a large bruise on his left arm and complaints of a headache. He had vomited and was lethargic. He was admitted for a third post-traumatic subdural haematoma, when it was revealed that adherence to the prescribed regimen of rFVIIa was poor. CT of the head revealed a small (3-4 mm) left-to-right midline shift and effacement of the anterior horn of the left lateral ventricle without hydrocephalus. Neurosurgery was consulted but surgery was ultimately not immediately pursued due to the patient's high risk of bleeding and because his neurological status was stable. His subdural haematoma was treated intensely with rFVIIa therapy 90 mcg/kg every 2-4 hours.

Given the patient's history of FIX inhibitors and the severity of his repeat intracranial haemorrhages, an inhibitor eradication regimen was designed for the patient based on previously published methods to improve the efficacy of the factor.⁵ A written informed consent was obtained from the patient's parents. Immediately prior to the start of factor desensitization, cyclophosphamide 600 mg/m² was administered along with three doses of mesna 240 mg/m² (day 0). Desensitization was initiated following cyclophosphamide administration (day 1). A FIX desensitization protocol was designed for the patient by hybridizing two published methods described by Dioun et al⁶ and Jamieson et al⁷ (Table 1). In brief, FIX was first administered at a starting dose of 0.00125 units/kg via slow IV push. Doses and infusion durations were incrementally increased for ten doses over the course of 6 hours until a desired cumulative dose of 60 units/kg was achieved. Premedications included dexamethasone 0.15 mg/kg every 8 hours for 3 days, ondansetron 0.15 mg/kg every 8 hours for 3 days, diphenhydramine 0.5 mg/kg one time,

and famotidine 0.4 mg/kg twice daily, all given intravenously. Following the desensitization, 60 unit/kg doses of FIX was administered daily as a continuous infusion for 2 days and then daily over 30 minutes thereafter for the remainder of his hospitalization (7 additional days). AlphaNine (plasma-derived FIX; Grifols), which is our institution's formulary FIX product, was the FIX product used in our desensitization protocol. Of note, AlphaNine has the volume of distribution of 1 with a half-life of 18 hours. By contrast, BeneFIX, when used in paediatric patients, has a volume of distribution of 1.4 and a half-life of 18 hours.

Three cycles of eradication and desensitization were administered to our patient (Figure 1). FIX challenge was not considered prior to eradication and desensitization due to the patient's history of multiple anaphylactic episodes to FIX even following diminished titre. The patient was negative for inhibitor 1 day after the first cycle of cyclophosphamide (day 1), which was expected due to his history of diminishing inhibitor titre following FIX withdrawal. The first cycle of desensitization and nine infusions of FIX infusions were tolerated without anaphylaxis and led to an improved neurological status with resolution of lethargy. The

patient was discharged with the intention of continuing daily FIX infusions.

One day after discharge, the patient had an anaphylactic event following his first ever exposure to peanut. Note that the anaphylaxis occurred shortly after peanut butter consumption and before his scheduled daily infusion of FIX. He was admitted for this episode of anaphylaxis at which point laboratory analysis revealed that he had experienced a reappearance of his FIX neutralizing antibody (21 BU). Challenge with FIX at that time did not result in an allergic reaction—supporting eradication of the FIX anaphylaxis associated antibody. The patient was discharged with rFVIIa and a scheduled follow-up for repeat desensitization.

Six days after confirmed reappearance of inhibitor, the patient was readmitted due to complaints of severe headache. The patient presented with drowsiness, clumsiness, slurred speech, decreased spontaneous movement of the left arm and an unsteady gait. Of note, the patient had experienced head trauma 3 days before he was brought to the clinic; furthermore, he had not received a single dose of rFVIIa (Note that this poor adherence is reflected in Figure 1, where there is a 5-day therapy-free interval). rFVIIa treatment was

TABLE 1 Desensitization protocol using AlphaNine (Grifols)

Day	AlphaNine dose (U/kg)	AlphaNine cumulative daily dose (U/kg)	Method	Interval from infusion of previous dose
1	0.00125	0.00125	Slow IV Push (2-5 min)	
	0.007	0.00825	Slow IV Push (2-5 min)	20 min
	0.0125	0.02075	Slow IV Push (2-5 min)	20 min
	0.07	0.09075	Slow IV Push (2-5 min)	20 min
	0.125	0.21575	Slow IV Push (2-5 min)	30 min
	0.7	0.91575	Intermittent Infusion over 30 min	30 min
	1.25	2.16575	Intermittent Infusion over 30 min	60 min
	7	9.16575	Intermittent Infusion over 30 min	60 min
	12.5	21.66575	Intermittent Infusion over 30 min	60 min
	32	53.66575	Intermittent Infusion over 30 min	60 min
2	60	60	Continuous Infusion over 24 h	24 h
3	60	60	Continuous Infusion over 24 h	24 h
4	60	60	Intermittent Infusion over 30 min	24 h
5	60	60	Intermittent Infusion over 30 min	24 h
6	60	60	Intermittent Infusion over 30 min	24 h
7	60	60	Intermittent Infusion over 30 min	24 h
8	60	60	Intermittent Infusion over 30 min	24 h
9	60	60	Intermittent Infusion over 30 min	24 h

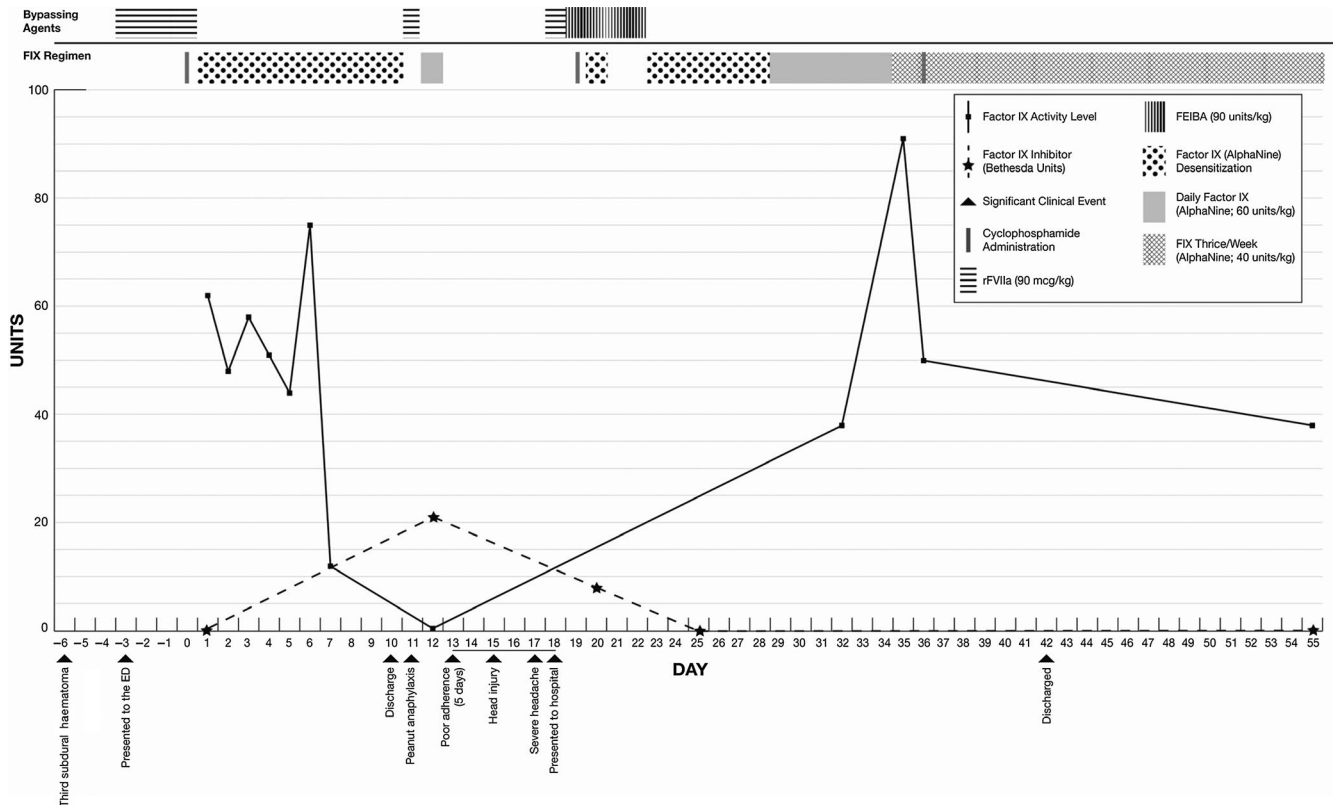


FIGURE 1 Course of the inhibitor activity, factor IX activity level, and clinical events

initiated for 1 day at 90 mcg/kg every 2–3 hours for eight doses and 100 mcg for one dose. The patient continued to have a severe headache and appeared disoriented. rFVIIa was switched to activated prothrombin complex concentrate (FEIBA[®], Shire) for 8 days in an attempt to improve patient response. FEIBA[®] was dosed at approximately 90 units/kg every 12 hours for 4 days and then every 24 hours for 4 days. Cycle two of immune eradication therapy and desensitization was commenced and again well tolerated. CT imaging showed improvement of acute haemorrhage. Headache and abnormal gait had resolved at time of discharge, and the patient was alert and oriented with an appropriate affect. FIX inhibitor level had been reduced to 8 BU 1 day after cycle two and was negative day 6 post cycle two. One month after the patient's third cycle of immune eradication therapy and desensitization, he was still negative for inhibitor. He has tolerated infusions of AlphaNine (40 units/kg) three times per week for 3 months following eradication of his inhibitor. This patient's inhibiting antibodies and episodes of anaphylaxis have resolved following FIX inhibitor immune eradication with cyclophosphamide. He has not experienced a CNS bleed 6 months after finishing his eradication and desensitization therapy.

Contributors to hypersensitivity and inhibitor development in PWHB include extravascular distribution of FIX, high concentrations of FIX protein in replacement therapy and complete gene deletions (such as the deletion seen in our patient).² Identifying the gene mutation in PWHB is an important step in understanding risk for inhibitor development and anaphylaxis and can guide physicians in management of these patients.²

In a 1997–2006 registry of patients with factor IX deficiency and a history of anaphylaxis or allergy to FIX concentrates, a total of 39 patients with inhibitors underwent an immune tolerance induction regimen with FIX.⁸ Only five total patients were successfully treated with an ITI modality that included FIX product only ($n = 25$; two successful treatments) or FIX product together with immunomodulation ($n = 14$; three successful treatments) using a combination of cyclophosphamide, γ -globulin and/or corticosteroids, with or without plasmapheresis. Rituximab has also been reported as an immunosuppressant in ITI protocols.⁹ Overall, the reported success of ITI induction in FIX deficiency with inhibitors is low and even when effective this treatment is associated with severe complications.^{3,10} Specifically, patients with a history of anaphylaxis to FIX concentrates (such as our patient) have a significant risk of developing nephrotic syndrome during ITI.¹⁰ The scarcity of inhibitor formation in PWHB offers few chances to explore treatment options and develops detailed and reliable treatment regimens for this subset of patients. High-dose cyclophosphamide with rapid tapering of factor VIII immune tolerance has previously been reported to be effective in the treatment of inhibitors in haemophilia A—especially in patients at high risk for immune tolerance failure.⁵ This report describes an effective eradication of FIX inhibitor and anaphylactic antibodies in a patient with severe haemophilia B using immune eradication with cyclophosphamide and factor IX desensitization. Our outcome indicates that immune eradication using cyclophosphamide and FIX desensitization may be an effective treatment in PWHB at high risk for immune tolerance failure.

Limitations: Follow-up monitoring of this patient post-FIX eradication was short (6 months).

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DISCLOSURES

NLK has acted as a paid consultant to Grifols and Octapharma—no conflict with this study.

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