

## CASE REPORT

# Achievement of immune tolerance in a patient with haemophilia B and inhibitory antibodies, complicated by an anaphylactoid reaction

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**Summary.** We describe a young boy with severe haemophilia B who developed inhibitory antibodies and an anaphylactoid reaction to factor IX. Immune tolerance was achieved by desensitisation with

escalating doses of factor IX followed by the Malmö regimen.

**Keywords:** haemophilia B, inhibitors, tolerance

The development of inhibitory antibodies in patients with haemophilia B is an uncommon event; arising in 3–5% of such patients. Treatment of these patients with immune tolerance induction tends to be difficult with fewer successes than haemophilia A (rates of 15–60% vs. 73–80% are quoted), as well as there being potential problems such as the development of nephrotic syndrome and/or systemic allergic reactions (anaphylactic or anaphylactoid) to factor IX (FIX) concentrates.

Here we shall discuss the case of a young boy with severe haemophilia B who developed both inhibitory antibodies to FIX and had an anaphylactoid reaction to treatment. He was successfully tolerized with a combination of desensitization therapy and the Malmö regimen.

A 3½-year-old boy was diagnosed at the age of 7½ months with severe haemophilia B (FIX <1%) to a family with no preceding history. He had presented with a 2-month history of easy bruising and two separate episodes of spontaneous haemarthrosis. Genetic testing later revealed the defect in the FIX gene to be a stop mutation: C31118T, R333X. This is known to be associated with severe haemophilia B but had not previously been reported in association with inhibitory antibody development.

He was treated with ‘on demand’ therapy and received only 4 days treatment with BENEFIX (recombinant FIX; Wyeth Europa Ltd, Maidenhead, UK) before routine testing showed a borderline positive result on an inhibitor screen [1]. At that time a formal Bethesda assay was negative. However, 1 month later, the inhibitor screen was unequivocally positive though the Bethesda titre (Nijmegen modification) was still negative (<0.3 BU mL<sup>-1</sup>).

Two weeks later he developed a spontaneous bleed into his left knee that required treatment. He received his 12th dose of BENEFIX for this bleed on the paediatric ward with full resuscitation facilities on hand, in view of his positive inhibitor screen. He suffered an acute systemic allergic reaction to the BENEFIX requiring oxygen, epinephrine, chlorphenamine and hydrocortisone as immediate treatment. The knee bleed responded to the BENEFIX but the inhibitor titre rose. On the day of this treatment it was documented as 1.6 BU mL<sup>-1</sup> and the level peaked to 10.3 BU mL<sup>-1</sup> 2 weeks later.

He went on to suffer recurrent haemarthroses, in particular to his left knee for which he received treatment with recombinant VIIa. Unfortunately, he developed a fixed flexion deformity to his knee and 6 months after the antibody was first detected an orthotic brace was fitted to try to reduce further joint damage.

Subsequent investigations revealed negative skin prick tests to 1:10 and neat solutions of BENEFIX and a negative intradermal test to a 1:100 dilution of BENEFIX. His total serum IgE was 3 KU L<sup>-1</sup> (NR

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0.5–23) and antiFIX antibodies of IgG or IgE isotype were undetectable by ELISA (assay details available on request). Hamster-specific IgE levels, tested on account of the presence of trace amounts of hamster protein in FIX were negative. In view of the negative immunological investigations, his allergic reaction was deemed to be anaphylactoid in origin.

It was clear that a definitive treatment strategy was required to minimize long-term problems from bleeding. After much deliberation, it was decided with the consent of his parents to give him desensitization therapy followed by the Malmö regimen (which combines immunosuppression with high doses of factor replacement therapy as a means of immune tolerance induction) (See Table 1). The combined aim was to prevent further anaphylactoid reactions and eliminate the inhibitory antibodies.

Prior to starting this treatment, 10 months after the antibody was first detected, the inhibitor level was  $<0.3$  BU mL<sup>-1</sup>. It rose to its highest postimmune tolerance level after week four of treatment (0.7 BU mL<sup>-1</sup>) but after 6 weeks the Bethesda assay was again negative ( $<0.3$  BU mL<sup>-1</sup>) and he had satisfactory FIX levels with once a day treatment. After 8 weeks he could be treated on alternate days

and after 10 weeks the inhibitor screen became negative. He now receives prophylactic BENEFIX administered at home every third day. At no time during this treatment did he develop proteinuria.

The development of inhibitory antibodies is one of the major complications of replacement therapy for patients with haemophilia. Added to this, in patients with haemophilia B there can be complications such as anaphylaxis or anaphylactoid reactions and the development of nephrotic syndrome (which can occur as late as 9 months into immune tolerance) [2].

The ideal treatment for patients with inhibitory antibodies is the induction of immune tolerance with the aim of eradicating the inhibitor. In haemophilia B this is less successful than in haemophilia A and, as mentioned, anaphylaxis can complicate this already difficult scenario. It is thought that anaphylaxis usually arises due to an IgE mediated allergic response to the exogenously administered FIX, though anaphylaxis due to IgG<sub>1</sub> has also been reported. Not all patients with inhibitors to FIX develop anaphylaxis and this association is seen more commonly in patients with undetectable FIX levels most frequently caused by either complete gene deletions or major abnormalities due to missing exons or stop codons. It has been hypothesized that those patients with anaphylaxis to FIX may also have a genetic predisposition for increased immunoglobulin isotype switching to IgE thus leading to their allergic response.

Desensitization is a well-practised method used to eliminate IgE mediated anaphylactoid reactions to allergens and indeed it has been reported in the literature for the elimination of anaphylactic reactions to FIX [3]. Another method of suppression of the anaphylaxis during immune tolerance therapy was reported by Barnes *et al.* using plasmapheresis and antigen exposure [4].

Here a specific protocol was devised for our patient that incorporated the use of the immunomodulators intravenous (i.v.) IgG and hydrocortisone prior to the stepwise incremental administration of FIX. In this case the desensitization programme worked well and enabled immune tolerance to be commenced.

Factor IX inhibitors are rare events and there is no clear consensus as to how to effectively treat them. A handful of reports have looked at various methods to induce tolerance, with varied success. These include immunoadsorption, low dose BENEFIX alone (40–50 IU kg<sup>-1</sup> dose<sup>-1</sup>), low dose followed by high dose BENEFIX (25 IU kg<sup>-1</sup> dose<sup>-1</sup> 3 × per week up to 220 IU kg<sup>-1</sup> dose<sup>-1</sup> alt. die.), the use of cyclosporine

**Table 1.** Protocol for immune tolerance induction.

| Day     | Immune tolerance regimen                                                                                                                                                |
|---------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| 1 and 2 | i.v. Ig 1 g kg <sup>-1</sup>                                                                                                                                            |
| 3       | Hydrocortisone 4 mg kg <sup>-1</sup> i.v.<br>Factor IX<br>0.1 U*<br>0.2 U*<br>0.4 U*<br>0.8 U*<br>1.0 U*<br>2.0 U*<br>4.0 U*<br>8.0 U*<br>16.0 U*<br>32.0 U*<br>64.0 U* |
| 4       | 100 U Factor IX                                                                                                                                                         |
| 5       | 400 U Factor IX                                                                                                                                                         |
| 6       | 800 U Factor IX                                                                                                                                                         |
| 7       | 1600 U Factor IX                                                                                                                                                        |
| 8 and 9 | Hydrocortisone 4 mg kg <sup>-1</sup> i.v.<br>Cyclophosphamide 12–15 mg kg <sup>-1</sup> i.v.<br>BENEFIX to maintain FIX 30–80 IU dL <sup>-1</sup>                       |
| 10      | Oral cyclophosphamide 2–3 mg kg <sup>-1</sup><br>BENEFIX to maintain FIX 30–80 IU dL <sup>-1</sup>                                                                      |
| 11–15   | Oral cyclophosphamide 2–3 mg kg <sup>-1</sup><br>i.v. IgG 0.4 g kg <sup>-1</sup><br>BENEFIX to maintain FIX 30–80 IU dL <sup>-1</sup>                                   |
| 16–19   | Oral cyclophosphamide 2–3 mg kg <sup>-1</sup><br>BENEFIX to maintain FIX 30–80 IU dL <sup>-1</sup>                                                                      |

\*Given by slow intravenous injection at 15 minute intervals.

and with more promising results the Malmö regime (references available on request).

The Malmö regimen has been used for induction of immune tolerance for both haemophilia A and B since 1982. It combines immunoadsorption (if necessary for removal of a high titre of inhibitory antibody), with oral cyclophosphamide and i.v. IgG as immunosuppression and FIX replacement. Success rates were given at 43% for patients with haemophilia B. Some of these patients required multiple treatments but eventually six of seven patients achieved immune tolerance [5]. The Malmö regime is reported to be more successful in those with a low peak inhibitor titre, relatively low historical peak titres and a long interval as the previous replacement therapy. Our patient fulfilled all three criteria. The parents were particularly keen on this approach as if successful we thought tolerance induction would take place rapidly reducing the chance of developing nephrotic syndrome.

This case report highlights the successful use of a two pronged approach to treat a complex clinical problem using desensitization and immune tolerance induction therapy. There are few patients with these problems reported in the literature; this approach may therefore be a potential method for managing these difficult individuals.

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## References

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