INHIBITORS IN HEMOPHILIA: A PRIMER

Fifth Edition

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INHIBITORS IN HEMOPHILIA: A PRIMER

What are inhibitors?

*Note: bolded terms are defined in the glossary at the end of this article.*

One way our immune system is designed to protect us from foreign things is by making antibodies. A person with hemophilia either produces no *clotting factor* (most cases of severe hemophilia) or an altered dysfunctional factor (most cases of mild/moderate hemophilia A [factor eight (FVIII) deficiency] and hemophilia B [factor nine (FIX) deficiency]). When such people are exposed to *factor concentrates* to replace the clotting factor (FVIII or FIX) that they are missing or have in an altered form, their immune system may see it as a foreign protein and develop neutralizing *allo-antibodies* called “inhibitors” against it. This then makes factor concentrate replacement ineffective for the treatment or prevention of bleeds. Inhibitor development is a much more common problem in people with hemophilia A than in those with hemophilia B.

Inhibitors present a significant management challenge for people with hemophilia (PWH). FVIII inhibitors bind to functional *epitopes* that are most commonly found in the A2, C1, and C2 domains of the factor protein. This binding interferes with the function of infused FVIII. FVIII inhibitors in patients with hemophilia A are mainly *immunoglobulin G* (IgG) antibodies of the IgG1 and IgG4 subclasses. IgG4 antibodies predominate in patients with high-titre inhibitors (HTI) while IgG1 antibodies are more abundant in patients with low-titre inhibitors (LTI). See the *Laboratory diagnosis* section below for a discussion of HTI and LTI inhibitors.

Not all immune responses to factor in hemophilia patients are inhibitors. Some patients can develop non-neutralizing antibodies. These are also IgG antibodies but as they do not target sites that are crucial to the activity of the factor, they do not inhibit or neutralize the coagulant function of factor. Still other patients (mainly those with severe hemophilia B) may develop *anaphylaxis*, an acute allergic immune reaction that may be caused by IgE antibodies.

Inhibitor antibodies to FVIII may also arise as *auto-antibodies* in people who do not have hemophilia; this is commonly referred to as acquired hemophilia. Such persons (who are not born with hemophilia) tend to be quite elderly and may develop antibodies that attack and destroy the FVIII which they produce due to a problem with their immune system. For more information on acquired hemophilia, the reader is encouraged to refer to No. 38 in the WFH Treatment of Hemophilia series, *Acquired Hemophilia* [1].

**Immune response to FVIII and FIX**

Why some people with hemophilia develop inhibitors and others don’t remains a mystery. Although we know that some patients are at higher risk of inhibitor development due to a combination of various genetic and environmental risk factors (discussed in the *Possible risk factors for inhibitor development* section below), ultimately it is still not known why one patient with a very similar risk profile for inhibitor development to another patient develops inhibitors, while the other does not. Immunologists continue to study inhibitor development in order to try to gain a better understanding of the process so that in the future we may potentially be able to prevent inhibitor development.

**Detecting inhibitors**

Inhibitors are usually detected in one of two ways. They may be discovered during routine inhibitor screening; or, alternatively, inhibitors may be suspected when a patient fails to respond to treatment with factor concentrates, meaning that replacement of the clotting factor no longer stops or prevents bleeding.
As it is better to detect inhibitors before being in a situation where a patient fails to respond to treatment, it is important to screen patients for inhibitor development particularly when they are at highest risk for developing them. The highest incidence of inhibitor development occurs during the first 20 exposure days (ED) to factor. This can occur when children are being treated on demand (i.e., episodically) or may occur after they have started prophylaxis. Such children should be screened frequently; many clinicians advise inhibitor testing every 5 ED until the patient reaches 20 ED, then every 10 ED until 50 ED are reached, and then at least twice per year until they reach 150 ED [2]. For children starting prophylaxis at an early age, most inhibitors, if they occur, will do so by 1 to 2 years of age. In situations where prophylaxis is not available and children are being treated on demand, it may take considerably longer to reach 50 ED. Close surveillance of clinical response to each infusion is important and inhibitor testing should be carried out if at all possible at any indication of a failure of response to factor.

Adults with more than 150 ED need less frequent screening for inhibitors. Inhibitor screening should be considered for all adults in certain instances such as following intensive exposure to factor, prior to undergoing major surgery, or when clinical response to treatment of bleeding is suboptimal. A second smaller peak of inhibitor development is seen in older age; the first and main peak is of course in very young children. It is also important to screen patients with mild/moderate hemophilia A after intense exposure, such as in surgery.

### Laboratory diagnosis

Inhibitors are detected and quantified by the functional Bethesda assay introduced in 1975, which relies on titration to measure inhibitors. In this assay, pooled normal plasma as the measure of FVIII or FIX is added to an equal amount of patient plasma. This test plasma sample, along with a control sample of buffered pooled normal plasma, is incubated at 37°C for 2 hours in the case of a FVIII assay and 10 minutes in the case of a FIX assay (FIX inhibitor kinetics differ from FVIII inhibitors in that the FIX antigen/antibody reaction reaches its peak level faster). At that point, a factor coagulation activity test is done to measure the residual FVIII/FIX level. If there are no inhibitors in the patient plasma then the FVIII or FIX in the pooled normal plasma of the test sample will be unaffected and the test results will reflect its activity. If, instead, there are inhibitors in the patient plasma, they will neutralize (essentially destroy) the factor in the normal pooled plasma. It is important to add a buffer to the pooled normal plasma of the control sample prior to incubation in order to correct for factor deterioration during incubation and improve FVIII/FIX stability and the specificity of the assay.

One Bethesda Unit (BU) is defined as the amount of inhibitor that neutralizes 50% of the FVIII/FIX in the test plasma sample as measured at the end of the 2-hour (FVIII) or 10-minute (FIX) incubation period. In the “Nijmegen” modification of the Bethesda test, introduced in 1995, the control consists of normal pooled plasma incubated with immune-depleted FVIII/FIX-deficient plasma buffered with imidazole to pH 7.4.

The strength of the inhibitory effect corresponds to the number of BU; the greater the number, the more inhibitors are present. As recommended by the International Society on Thrombosis and Haemostasis (ISTH), the cut-off value for what constitutes the presence of inhibitors is defined as a titre ≥0.6 BU using the Nijmegen modification of the Bethesda assay documented on 2 separate occasions, usually within a 4-week period [3].

The Bethesda assay differentiates low-titre inhibitors from high-titre inhibitors; the former are generally defined as having an inhibitor titre <5 BU while the latter are defined as occurring when the inhibitor titre ≥5 BU. The Bethesda assay will not, by definition, detect non-neutralizing antibodies (i.e., antibodies to FVIII or FIX that do not inhibit the coagulation function of these proteins). However, both neutralizing and non-neutralizing antibodies to FVIII or FIX may be detected by an enzyme-linked immunosorbent assay (ELISA) or fluorescence-linked immunoassay. The ELISA assay detects and quantifies all antibodies present, whether or not they are inhibitory, however the clinical significance of this assay is still under discussion.

An inhibitor reduces both factor recovery (a reflection of how the patient responds to factor infusion) as well as factor half-life (a reflection of how rapidly the factor is degraded). Often once inhibitors are detected, particularly in the case of LTIs, patients undergo some type of
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pharmacokinetic evaluation in order to determine the impact of the inhibitor neutralizing factor.

High-titer and low-titre inhibitors behave differently and consequently are managed differently (see the sections below: What to do when low-titre inhibitors develop and What to do when high-titre inhibitors develop). For example, patients with high-titre inhibitors may experience a drop in their inhibitor titre if they are not exposed to factor, however they will usually develop a strong anamnestic response to FVIII with a rise in inhibitor titre if they are subsequently re-exposed to FVIII, while under the same conditions patients with low-titre inhibitors will not.

Incidence and prevalence

Inhibitors are most commonly encountered in people with severe hemophilia A (overall 25-40% lifetime risk) compared to those with moderate/mild hemophilia A (overall 5-15% lifetime risk). It should be noted that whereas most mutations that cause moderate/mild hemophilia A have a very low risk of inhibitor development (<5%), some carry a much higher risk of inhibitor development (up to 15%) [4]. For patients with hemophilia B, the risk of inhibitor development is about 1-5% and is almost only seen in patients with severe hemophilia B in whom the hemophilia is caused by null mutations – defined as mutations that result in no factor being produced. In most cases in hemophilia B, these null mutations are large deletions and nonsense mutations.

In patients with severe hemophilia (A or B), the risk of inhibitor development is highest during the first 20 exposures to factor replacement after which the risk decreases dramatically, particularly from 20 ED to 50 ED. After 50 ED the risk, although already quite low, decreases further reaching a very low steady-state rate of 2-5 per 1,000 patients per year by 150 ED [5]. Therefore, having reached a minimum of 150 ED has been the classical definition of a patient referred to as a previously treated patient (PTP).

In people with moderate/mild hemophilia A, if inhibitors develop, they do so on average at a much older age and often following intensive FVIII exposure, such as occurs in the setting of surgery [7]. Inhibitors that develop in people with moderate/mild hemophilia often behave differently than inhibitors in people with severe hemophilia and more like what is seen in acquired hemophilia.

The incidence of inhibitor development is often expressed as “all inhibitor” development or as HTI development. All inhibitors include both HTIs and LTIs; the latter are usually considered as transient although a significant proportion can subsequently convert to HTIs [8]. If inhibitor testing is performed frequently then more transient LTIs will be detected. Consequently, with more frequent inhibitor testing over the last several decades, the rate of all inhibitor development has risen whereas the rate of HTIs has remained fairly constant. There may be additional reasons for an observed increase in inhibitor detection in the last 2 decades.

The number of people with inhibitors present in a population at any given time (referred to as prevalence) reflects a number of elements: the incidence of inhibitor development, the spontaneous disappearance of transient LTIs, the active eradication of inhibitors through immune tolerance induction (ITI) therapy (discussed below), and deaths among patients with inhibitors. In countries with good availability of bypassing agents, inhibitor patient deaths are an infrequent event and would be the element with the least impact on inhibitor prevalence. As such, the prevalence of inhibitors is much lower than the incidence: in the case of severe hemophilia A, the prevalence of inhibitors is approximately 5-10%, meaning that at any given time 5-10% of people with severe hemophilia A will have inhibitors.

Acquired hemophilia is uncommon and estimated to occur in 1.4 persons per million people per year. FIX auto-antibodies are even rarer still.

Possible risk factors for inhibitor development

A number of factors influence the risk of inhibitor formation. These can be categorized into non-modifiable genetic factors and potentially modifiable environmental factors (Table 1).
### TABLE 1: Non-modifiable and potentially modifiable risk factors for inhibitor development

<table>
<thead>
<tr>
<th>Risk factors</th>
<th>Summary</th>
<th>Level of support</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Non-modifiable genetic risk factors</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>F8 mutation type (null vs. non-null and position of mutation) [9]</td>
<td>Type of mutation</td>
<td>Risk of inhibitor development</td>
</tr>
<tr>
<td>Null</td>
<td>Multi-domain deletions</td>
<td>≈75%</td>
</tr>
<tr>
<td></td>
<td>Light chain nonsense mutations</td>
<td>30-40%</td>
</tr>
<tr>
<td></td>
<td>Intron 22 inversion</td>
<td>20-25%</td>
</tr>
<tr>
<td></td>
<td>Single domain deletions</td>
<td>15-25%</td>
</tr>
<tr>
<td></td>
<td>Small non-A run insertions/deletions</td>
<td>15-20%</td>
</tr>
<tr>
<td></td>
<td>Heavy chain nonsense mutations</td>
<td>10-20%</td>
</tr>
<tr>
<td>Non-null</td>
<td>FVIII missense mutations</td>
<td>&lt;10%</td>
</tr>
<tr>
<td></td>
<td>Small A run insertions/deletions</td>
<td>&lt;5%</td>
</tr>
<tr>
<td></td>
<td>Splicing mutations</td>
<td>&lt;5%</td>
</tr>
<tr>
<td><strong>Note:</strong> Nonsense mutations cause premature stop codons. Missense mutations</td>
<td></td>
<td></td>
</tr>
<tr>
<td>allow a full-length protein to be made and circulated although it has a mistake</td>
<td></td>
<td></td>
</tr>
<tr>
<td>in it. A few specific missense mutations are associated with a high frequency</td>
<td></td>
<td></td>
</tr>
<tr>
<td>of inhibitors.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Family history [10]</td>
<td>3.2-fold higher risk (95% CI 2.1-4.9) if there is a family member with inhibitors</td>
<td>Well established</td>
</tr>
<tr>
<td>Ethnicity [11]</td>
<td>1.9-4.7-fold higher risk in non-Caucasians (Black African descent &gt; Latin American &gt; Caucasian)</td>
<td>Established but not well understood</td>
</tr>
<tr>
<td>TNF-α [12]</td>
<td>IL-10: allele 134 increases risk</td>
<td></td>
</tr>
<tr>
<td>IL-10 [13]</td>
<td>TNF-α: -308 A/A increases risk</td>
<td></td>
</tr>
<tr>
<td>CTLA-4 polymorphisms [14]</td>
<td>CTLA-4: T-allele decreases risk</td>
<td></td>
</tr>
<tr>
<td>F8 haplotypes [15]</td>
<td>H3 or H4 haplotypes have higher risk of inhibitors as current FVIII treatment products mainly consist of H1 and H2 haplotypes</td>
<td>Conflicting reports</td>
</tr>
<tr>
<td>MHC gene classes I/II or HLA polymorphisms [15]</td>
<td>2-fold higher risk for HLADR15 and HLA-DQ6 and inhibitor development</td>
<td>Conflicting reports</td>
</tr>
<tr>
<td><strong>Potentially modifiable environmental risk factors</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Trauma/surgery [16]</td>
<td>Major surgical procedures and trauma leading to peak treatment moments</td>
<td>Established but not well understood</td>
</tr>
<tr>
<td></td>
<td>increase risk of inhibitor development</td>
<td></td>
</tr>
<tr>
<td>Inflammation/infection [17]</td>
<td>May increase inhibitor development</td>
<td>Established but not well understood</td>
</tr>
<tr>
<td>Intense exposure, especially early in life [16]</td>
<td>Increases risk of inhibitor development</td>
<td>Established but not well understood</td>
</tr>
<tr>
<td>Factor concentrate type [18]</td>
<td>Some evidence to suggest that conventional recombinant factor conveys higher risk of inhibitors than plasma-derived (see below)</td>
<td>Conflicting reports</td>
</tr>
<tr>
<td>Early initiation of prophylaxis [19]</td>
<td>May confer some protection</td>
<td>No clear evidence</td>
</tr>
</tbody>
</table>

**Note:** Age at first exposure and vaccinations were considered, at one point, to be potential environmental risk factors but now are no longer considered to be factors that increase the risk of inhibitor development [20].
Of the above genetic risk factors, the most predictive of inhibitor development are genetic mutation, family history of inhibitors, and ethnicity. In contrast, the predictive value of \( F_8 \) haplotype, MHC gene class/HLA polymorphisms, and immune regulatory gene polymorphisms are fairly weak.

### Environmental risk factors

Environmental factors that have been suggested to influence the risk of developing inhibitors include both treatment related (i.e., type of product, dosing regimen) and immune system activating risk factors (so-called “danger” signals – a term that refers to the release of inflammatory substances from damaged tissue) [17]. Intense exposure, also referred to as “peak” treatment moments, defined as episodes in which factor is infused at least once per day for 3 consecutive days, have been shown to be related to increased risk of inhibitor formation. Taking these into account, clinicians speculate that it might be possible to reduce a patient’s risk of inhibitor development through measures such as postponement of elective surgery in order to prevent intense exposure, avoidance of excessive treatment for relatively minor bleeding episodes in very young children who have not yet reached 50 ED to FVIII or FIX, and avoidance of large doses of factor when children are very young and, as such, at highest risk of inhibitor development. Starting prophylaxis early may also reduce the risk of inhibitors, although this remains controversial.

### Effect of replacement factor type

While there is no question that all factor concentrates carry a risk of inhibitor development, the question of whether concentrate type (plasma-derived or recombinant) has a role in inhibitor development has been a topic of debate since the introduction of recombinant factor concentrates in the 1990s. Until recently, there had been no randomized trials comparing inhibitor incidence in PUPs with severe hemophilia A receiving either recombinant FVIII concentrates (rFVIII) or plasma-derived concentrates. However, in the Survey of Inhibitors in Plasma-Product Exposed Toddlers (SIPPET) study, investigators conducted a prospective randomized study and showed a significantly higher inhibitor rate in the group of PUPs receiving 1 of 4 recombinant FVIII concentrates compared to those receiving 1 of 4 plasma-derived FVIII/von Willebrand factor (VWF) containing concentrates (44.5% vs. 26.7% for all inhibitors, 28.4% vs. 18.5% for HTIs) [18]. Several newer recombinant products have not been studied in this way, nor have any high-purity plasma-derived products; therefore, no conclusions can be made about their relative risks. However, prompted by the SIPPET study, the European Medicines Agency (EMA) through its Committee for Medicinal Products for Human Use (CHMP) conducted a review of available data and released a statement in September 2017 concluding that there is no clear and consistent evidence of a difference in the incidence of inhibitor development between the two classes of FVIII: plasma-derived and recombinant [21].

Another ongoing debate has been whether all recombinant FVIII concentrates carry the same risk of inhibitor development or are some more likely to cause inhibitors than others based on such factors as differences in glycosylation and sulfation. When B-domain deleted rFVIII was introduced, there was concern that this concentrate posed a higher risk of inhibitor development than full-length factor concentrates. Regulators at both the U.S. Food and Drug Administration (FDA) and the European Medicines Agency have determined that there is no conclusive evidence supporting these concerns. In studies involving PTPs switching between different recombinant factor concentrates, no evidence has been found of increased inhibitor development [22]. Recent studies, including the large prospective cohort RODIN study, found that the incidence of inhibitor development was higher with one second generation full-length BHK rFVIII concentrate versus a third generation full-length CHO rFVIII concentrate (hazard ratio 1.6-1.75) [6]. This finding was also seen in several other large European studies [23, 24, 25]. Yet some epidemiologists and clinicians have voiced concerns with the methodology of such studies and consequently it is not, as yet, possible to draw definitive conclusions from such studies to declare any particular rFVIII to be more or less likely to lead to inhibitor development.

Many new FVIII and FIX concentrates have recently been licensed or are in different stages of development for clinical use, and some of these products have been designed with the intention of reducing inhibitor risk. In studies of PTPs switching to these new recombinant factor concentrates [both human cell line-derived and extended half-life (EHL)], very few inhibitors have so far been reported, which is generally the case for PTPs. Studies in PUPs are
currently underway. There are some theoretical reasons to suspect that the immunogenicity of these new factor concentrates may be less than that of established concentrates: the PEG or Fc moieties that characterize the EHL products could somehow shield the recombinant FVIII/FIX from the immune system leading to less inhibitor development, while a human cell-line derived FVIII/FIX may more closely mimic natural human FVIII leading to less inhibitor development. Of course, all of this remains speculative and the results of studies of the immunogenicity of new factor concentrates in PUPs are eagerly awaited; the reader is advised to consult the most recent literature.

Finally, it should be emphasized that a risk of inhibitors exists with all factor concentrates but it is better to accept that risk and treat bleeds than to avoid treating altogether.

Basic principles of management

When inhibitors are initially detected, a management plan needs to be quickly put into place for optimal care of the patient. This is best done in a hemophilia treatment centre experienced in the management of patients with inhibitors.

The first thing that needs to be done is to determine the inhibitor titre and classify the inhibitor (LTI or HTI) as the subsequent management will depend greatly on this. As previously mentioned, an LTI can progress to an HTI and if this occurs then management is generally as per HTI.

What to do when low-titre inhibitors (LTI) develop

It is important to realize that many LTIs may be transient, disappearing spontaneously without specific management within 6 months of initial documentation; despite continued factor exposure. For such patients, there may not be any need to attempt to eradicate the inhibitors (discussed below); consequently, most clinicians would recommend not initially changing therapy when patients develop LTIs. However, such patients need to be closely monitored with Bethesda assays every 2-4 weeks as their inhibitors can convert to HTIs. Furthermore, if such inhibitors persist for a long time or if the patient starts to experience recurrent bleeding, then there may be a role for immune tolerance induction. ITI is defined as a process by which the immune system is trained to better accept treatment with the missing clotting factor without producing further antibodies. For more details see the Immune tolerance induction (ITI) therapy section below.

Alternatively, porcine recombinant FVIII, available in some countries for the management of acquired hemophilia A, may also become available for patients with hemophilia A and LTIs. Porcine rFVIII is a recombinant B-domain deleted form of FVIII that is typically not as quickly inactivated or destroyed as human FVIII is when administered to patients with inhibitors. Porcine rFVIII may better evade inhibitors and, therefore, it might be possible to use it to treat bleeds in people with inhibitors, particularly if the inhibitor titre is not very high. An initial dose of 200 IU/kg of porcine rFVIII is recommended, with further dosing depending on plasma FVIII levels or clinical response. Some patients develop antibodies to porcine rFVIII after several days of treatment or after several episodes of treatment, becoming no longer responsive.

In the rare event that patients with mild hemophilia A develop LTIs displaying type 2 kinetics (i.e., inhibitors that do not totally inactivate endogenous FVIII) then desmopressin (DDAVP) may be sufficient to release enough FVIII to neutralize the circulating inhibitors and raise the plasma FVIII level enough to stop minor bleeding or allow minor surgical procedures.

What to do when high-titre inhibitors (HTI) develop

High-titre inhibitors tend to be persistent and they render the patient completely resistant to factor concentrates. Consequently, they demand significant alterations in the
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The management of a patient with the use of **bypassing agents**, recombinant FVIIa (rFVIIa), or activated prothrombin complex concentrates (aPCC), in order to treat and prevent bleeds (discussed in detail in the Bypassing agents section below).

In general, the first thing to be done once a hemophilia A patient develops HTIs is to avoid further FVIII exposure until ITI is commenced. Avoiding ongoing FVIII exposure will cause the person’s immune system to be less stimulated and produce less inhibitors. This causes inhibitor titres to fall. At this stage in treatment (prior to commencing ITI), treating bleeds and preventing bleeding during surgical procedures is best accomplished with rFVIIa since aPCC (Factor Eight Inhibitor Bypassing Activity, FEIBA®) contains small amounts of FVIII which may contribute to an anamnestic response; reported to occur in approximately 30% of FVIII inhibitor patients receiving FEIBA® [26].

The development of HTIs, particularly in young children, has often resulted in a central venous catheter (usually a port-a-catheter), if not already inserted, being inserted given the increased need for reliable venous access both for treating bleeds and to facilitate ITI. Once an inhibitor is confirmed, if peripheral venous access is deemed inadequate for ITI and management of intercurrent bleeds in such patients then port placement should be expedited following which ITI should be commenced.

A decision regarding how to treat bleeds in patients with HTIs depends on the inhibitor level, the severity of bleeding, and the patient’s previous therapeutic response.

Minor bleeding in patients with inhibitors may still be effectively controlled with **local hemostatic measures**, such as application of pressure for nosebleeds and anti-fibrinolytic therapy, such as tranexamic acid or epsilon aminocaproic acid. When such measures fail, or are deemed inadequate for the type of bleeding, then bypassing agents are generally required.

**Bypassing agents**

There are 2 types of bypassing agents: plasma-derived activated prothrombin complex concentrates (aPCC; the only commercially available one is FEIBA®) and recombinant FVIIa (rFVIIa). aPCCs are plasma-derived but virally attenuated, and contain prothrombin complex zymogens FII, FVII, FIX, and FX as well as small amounts of their activated forms (IIa, IXa, Xa, and especially VIIa) that stimulate the formation of a clot and stop bleeding, thus bypassing the requirement for FVIII or FIX. Both agents are reported to be effective in treating 90% of musculoskeletal bleeds and can be used for both major and minor surgical prophylaxis.

A comparison of the characteristics of the two agents is provided in Table 2.

**TABLE 2: Advantages and disadvantages of the two available bypassing agents**

<table>
<thead>
<tr>
<th>Typical regimen to treat bleeds</th>
<th>Advantages</th>
<th>Disadvantages</th>
</tr>
</thead>
</table>
| **FEIBA®** 50-100 IU/kg every 6-12 hours (max. 200 IU/kg/day) | • Lasts longer (vs. rFVIIa)  
• Can be given every 6-12 hours | • Plasma-derived  
• Large volume  
• 30-45 minutes to administer  
• Not to be given with tranexamic acid  
• Contains some FVIII  
• Has a higher rate of thrombosis if given concomitantly at high doses for >1 day in patients on emicizumab [27] (See section on non-factor therapies including emicizumab) |
| **rFVIIa** 2-3 doses of 90 μg/kg every 2-3 hours or Single dose of 270 μg/kg | • Recombinant  
• Small volume  
• Can be given over 2-5 minutes  
• Can be given with tranexamic acid  
• Appears to be safer when given in combination with emicizumab | • Does not last very long  
• Needs to be given more frequently |
Comparative studies have shown that the clinical efficacy of a single dose of aPCC (50-100 IU/kg) is essentially equivalent to that of 2 doses of rFVIIa (90-120 μg/kg) for treating joint bleeding [14]. Notably, however, some patients respond better to one agent than the other, highlighting the need to individualize therapy. As well, patient responses to either agent may vary over time and with the type of bleed. Where possible, patients should have a supply of one of these bypassing agents at home, allowing early home infusion at the first sign of bleeding (ideally within the first 2 hours). Some patients who are very experienced with using these bypassing agents may even have both at home, using one for either a specific type of bleed or for prophylaxis (see below), while the other may be used for other types of bleeds or for on-demand treatment of breakthrough bleeds. Failure to respond to one bypassing agent should prompt consideration of a switch to the other, which might necessitate hospitalization. In rare instances, the bleeding may be refractory to either, and use of both agents in sequential fashion may be appropriate in this context; however, close monitoring for thrombosis or consumptive coagulopathy (also referred to as disseminated intravascular coagulation) is required. Combination treatment should be used only in centres with extensive experience in managing patients with inhibitors.

When comparing the two available bypassing agents, it is clear that both have advantages and disadvantages versus the other (see Table 2). However, both bypassing agents are far less effective in treating and preventing bleeds than conventional factor concentrates used in patients without inhibitors. Furthermore, bypassing agents are less convenient to use due to their short half-life and the consequent need for frequent infusions (particularly with rFVIIa) or due to the need for prolonged infusion times (FEIBA®). Thrombosis, particularly deep vein thrombosis and myocardial infarction, have also been reported with both aPCC and rFVIIa [28]. Finally, unlike standard factor replacement therapy, in which factor assays can be used to guide therapy, there is no standardized laboratory test to quantify the activity of a bypassing agent in vivo. Therefore, apart from indirect measurements of overall hemostatic potential (such as thromboelastography [TEG] or a thrombin generation assay [TGA]), assessment of response to a bypassing agent must be based on clinical symptoms (i.e., ongoing pain, swelling, limited range of motion).

The use of bypassing agents for prophylaxis has typically been reserved for patients with a high bleeding tendency or pre-existing significant joint damage. However, there is increasing evidence to support the use of prophylaxis in patients with inhibitors to prevent bleeds and maintain function or limit deterioration in musculoskeletal status associated with bleeding into muscles and joints. Either rFVIIa (e.g., 90 or 270 μg/kg daily) or aPCC (e.g. 75-100 IU/kg 3-4 times weekly) can be used alone or in combination with standard ITI therapy. In patients undergoing ITI, once there is measurable factor recovery, prophylactic bypassing agents should be discontinued due to the risk of thrombosis when given in conjunction with high doses of factor. Breakthrough bleeds in patients on bypassing agent prophylaxis can be managed initially with additional doses of the same bypassing agent or with the alternate agent.

Non-factor therapies for patients with inhibitors (emicizumab and rebalancing agents)

In 2017, the results of the first clinical trial on the use of a non-factor therapy (emicizumab) in hemophilia A patients with inhibitors was published in the New England Journal of Medicine [27]. Emicizumab is a bispecific monoclonal antibody initially developed by researchers in Japan [29]. It was developed to mimic the activity of FVIII. Like FVIII, emicizumab brings FIXa and FX together, allowing the activation of FX which then allows the coagulation cascade to continue, ultimately leading to the production of a clot. Although emicizumab mimics the biological effect of FVIII, it is not FVIII and as such is not affected by anti-FVIII antibodies. A phase 3 trial of emicizumab, administered subcutaneously once per week in patients with hemophilia A and inhibitors, showed a significantly reduced (87%) rate of bleeding compared to patients treated with bypassing agents on demand. Based on these results, emicizumab was approved for use in hemophilia A patients with inhibitors in late 2017 by the FDA and other jurisdictions began granting market authorization in 2018. Overall, the results of using emicizumab in patients with hemophilia A and inhibitors are very encouraging. Nevertheless, caution is warranted when considering new non-factor therapies for hemophilia; several patients on emicizumab did show thrombotic complications.
including thrombotic microangiopathy (TMA) and there have been several reports of deaths in patients – although none have, as yet, been attributed to emicizumab, at the time of writing of this monograph (Summer, 2018). Emicizumab now raises a number of issues which the hemophilia community has not faced in the past: should some patients continue on bypassing agent prophylaxis rather than prophylaxis with emicizumab; should ITI still be attempted to eradicate inhibitors; should rescue ITI still be attempted; how should bleeds be treated in patients on emicizumab? These questions are being addressed in this developing story and the reader is advised to consult the most recent literature and regulatory guidance.

Other non-factor therapies are also in various stages of development. These molecules are designed to substitute for FVIII in the clotting cascade, but are completely different to FVIII.

A number of drugs which work to rebalance the equilibrium between bleeding and clotting by decreasing anti-coagulants that naturally occur in the human system (i.e., tissue factor pathway inhibitor [TFPI], anti-thrombin) are also in different phases of development. Of these, fitusiran (a molecule that works by reducing the production of anti-thrombin – a potent natural anti-coagulant – to improve the coagulation equilibrium) is the furthest along [30]. Although quite effective in reducing rates of bleeding, a death from a severe intracranial blood clot led to the manufacturer halting its use. While studies on fitusiran have recommenced, the future of this product is still not clear.

Since these types of drugs differ completely from FVIII or FIX replacement therapy, such agents hold the additional promise that they may be used in both hemophilia A patients with inhibitors to FVIII and hemophilia B patients with inhibitors to FIX. For hemophilia patients with inhibitors, these non-factor replacements, most of which can be given subcutaneously, offer much promise to improve quality of life. However as noted above, these drugs do carry risks (both known and unknown), and as such their ultimate clinical use is not guaranteed.

As the development of these novel agents is rapidly evolving, the reader is advised to consult the most recent literature and regulatory guidance for their current status.

Inhibitor eradication

Although there are several available therapeutic options for treating and preventing bleeds in people with hemophilia and inhibitors, none have been able to guarantee as good an outcome as specific FVIII or FIX treatment in non-inhibitor patients. With the advent of emicizumab, and as similar treatments are developed, this may no longer be the case in the future. Consequently, up until now people with inhibitors have generally experienced more frequent bleeding, including life-threatening bleeds, and have had greater disability in their day-to-day lives than people with hemophilia who do not have inhibitors [31]. In a recent Universal Data Collection (UDC) surveillance study by the U.S. Centers for Disease Control (CDC), it was shown that patients with persistent inhibitors have a higher rate of early death and worse quality of life [32]. Therefore, to date, for most individuals who develop HTIs, eradication of the inhibitors remains the best option.

Lowering inhibitor levels

Plasmapheresis, a method of removing plasma from the body by withdrawing blood, separating it into plasma and cells, removing the plasma (which contains antibodies), and transfusing the cells back into the bloodstream, may be a short-term option in treatment centres with the relevant expertise to lower inhibitor titres in patients who are not responding to bypassing agents or when bypassing agents are not available. Even in such centres, it is generally advocated only in cases of life-threatening bleeding. Plasmapheresis can remove much of the inhibitor, thus possibly permitting the short-term use of conventional factor replacement. However, this is only a temporary measure, since giving the factor will then stimulate the body to make large amounts of new antibody within several days. If time permits, (i.e., before an urgent but non-emergency surgical operation), plasmapheresis may be performed on 2 or 3 consecutive days.

Immune tolerance induction (ITI) therapy

If not treated with replacement factor concentrate for a long period, high-titre inhibitor levels may fall or even become undetectable. Yet when such patients are re-exposed to the specific factor concentrate, there will be an anamnestic response in 3-5 days, precluding further use of conventional factor. For such patients, ITI is the only therapeutic strategy with the potential to eradicate persistent FVIII or FIX inhibitors and restore normal factor
pharmacokinetics. ITI is comprised of regular (daily or several times weekly) infusions of variable doses of FVIII or FIX, administered for periods of months to years in an effort to tolerize the immune system to FVIII or FIX (i.e., to train the immune system to accept treatment with the missing clotting factor without producing antibodies).

The optimal protocol (combination of dose and frequency) for ITI has yet to be established; to date most experience has been garnered with inhibitors to FVIII. Doses as low as 25 IU/kg per infusion and as high as 300 IU/kg per infusion have been administered anywhere from 3 times/week to twice/day. It has been shown that patients with good prognostic features (mainly a historical peak inhibitor titre <200 BU and pre-ITI titre <10 BU) can achieve similar rates of inhibitor eradication success either with a very high-dose regimen (e.g., ≥200 IU/kg/day) or with a low-dose regimen (e.g. 25–50 IU/kg 3 times/week) [33]. The high-dose regimen has the advantage of less bleeding episodes and shorter time to tolerization, however it comes at a much higher cost. Evidence suggests that patients with poor prognostic factors benefit more from high-dose regimens.

Low-dose ITI regimens avoid the inconvenience of daily doses of factor (and the likely need for a port-a-catheter) and are less costly, thus they may offer a practical and effective approach to ITI in the context of significant resource constraints where large amounts of factor concentrate are not readily available. Low-dose ITI may also be of value for patients with persistent LTIs who start to experience recurrent bleeding.

The ideal product type for ITI of a person with hemophilia A and inhibitors (regular or extended half-life rFVIII, or high purity or plasma-derived FVIII/VWF) has been the subject of intense debate. Some clinicians believe that ITI outcomes with plasma-derived FVIII products that contain VWF are better and attribute this to the VWF shielding parts of the FVIII from recognition by the immune system. However, there is no definitive evidence regarding this matter. The current approach used in most treatment centres is to start ITI with the same product on which the inhibitors developed, although this is not based on any data either. If the response to initial ITI with a conventional rFVIII is suboptimal, then switching the ITI therapy to a plasma-derived VWF-containing FVIII, or more recently available EHL rFVIII, may be considered, where these products are available. Alternatively, some clinicians may choose to immediately start ITI with a plasma-derived FVIII/VWF concentrate or an EHL rFVIII concentrate.

Information collected through both ITI registries and prospective studies [34] allow for a better understanding of prognostic markers of ITI success or failure. These include low historical peak inhibitor titre and low peak titre during tolerization. Age at the start of ITI, ITI interruptions, as well as the length of time between inhibitor appearance and the start of ITI may all also be important in predicting success.

When to start ITI has also been controversial. Registry data from the 1990s showed that patients were more likely to achieve ITI success if they started with an inhibitor titre of <10 BU [35]; many clinicians interpreted this as an indication to wait until inhibitor titres dropped to <10 BU before commencing ITI. However, it should be noted that patients in registries commencing ITI with an inhibitor titre <10 BU consisted of 2 groups of patients; a group in whom the inhibitor titre had never risen to >10 BU (a very good prognostic group), along with a second group in whom the inhibitor titre had risen to >10 BU but in whom clinicians waited to commence ITI to allow the inhibitor titre to drop (a less favourable group). By combining these 2 types of inhibitor patients, it is possible that an erroneous conclusion may have been formulated – to wait to commence ITI until inhibitor titres drop to <10 BU.

An increasing number of centres are now commencing ITI as soon as possible without waiting for inhibitor titres to drop and several reports suggest very good results with this approach [36].

Successful ITI is defined by both the absence of residual antibody (a negative Bethesda titre, usually defined as a Bethesda titer of <0.6 BU), and by a return to normal factor pharmacokinetics (i.e., normal distribution and metabolism of factor when administered to the patient). Defining ITI failure is more problematic. Generally, failure to achieve ITI success after a certain amount of time (2-3 years) is usually used as the definition of failure, although some studies have also used failure of the inhibitor titre to drop a certain amount over a certain amount of time.

ITI, although time-consuming and costly, is effective in 60-80% of patients in which it is undertaken and
Inhibitors in Hemophilia: A Primer

consequently it is considered to be the standard of care in the case of high-titre inhibitor development in people with severe hemophilia. The success of ITI appears to be less pronounced in patients with mild/moderate hemophilia. Due to its high cost and the requirement for access to large quantities of factor concentrate, it is not always possible to undertake ITI in countries with significant resource constraints. Successful ITI has several benefits: it enables regular treatment with factor products including regular prophylaxis, increases quality of life, and, despite a very high short-term cost, reduces the cost of future care. Most of the experience with ITI derives from studies conducted in children. It is generally accepted that the risk of ITI failure is much greater in adults with longstanding HTIs, although there are some case reports of successful ITI in adults. The cost is, of course, also much greater in adults than in children due to the higher weight of adults which requires higher doses.

With the advent of emicizumab, which can be given to hemophilia A patients with FVIII inhibitors subcutaneously once per week, or potentially even less frequently, the need to eradicate inhibitors from an individual patient may be somewhat less pressing. However, patients with inhibitors on emicizumab will likely still need episodic treatment with bypassing agents should they experience a bleed or undergo surgery. Bypassing agents, when given to treat bleeds or manage surgery, are in general less convenient and (in the context of emicizumab use) less safe than the use of FVIII concentrates in non-inhibitor patients whether or not they are taking emicizumab. However, if patients with inhibitors are successfully tolerized then they could potentially simply use replacement FVIII to treat bleeds or for surgery even if they remain on emicizumab for long-term prophylaxis. The use of FVIII to treat bleeds or to undergo surgery has so far been found to be safe and effective when given to patients without inhibitors who are taking emicizumab.

Therefore, overall, at present there is still strong support for recommending ITI when a patient develops inhibitors. However, for patients who fail initial ITI, the support for trying rescue ITI may be diminished with the availability of emicizumab.

Also, whether non-factor therapies such as emicizumab will impact ITI regimens still requires investigation. Low-dose ITI regimens, which are much less expensive and much less of a burden, have been shown to take longer to achieve tolerance and to be associated with more bleeding than high-dose ITI regimens. With non-factor therapies such as fitusiran and emicizumab, perhaps patients/clinicians may choose a low-dose ITI regimen given concomitantly with a non-factor therapy; the purpose of the latter being to reduce bleeding while the purpose of low-dose ITI is to simply eradicate the inhibitors. The ramifications of these newer therapies on ITI remain to be determined.

**FIX inhibitors**

Inhibitor development is much less common in patients with hemophilia B, therefore, most clinicians are less experienced at managing such patients. FIX inhibitors are primarily antibodies of the IgG4 isotype, although some are of the IgG2 isotype. Most FIX inhibitors occur in individuals with large or complete deletions of the F9 gene and their development is often associated with severe allergic reactions, including anaphylaxis, to FIX administration. As anaphylactic reactions to FIX may occur very early on, it is recommended that the first 10 FIX exposures (in those with non-null mutations) to 20 FIX exposures (in those with null mutations) be given in a clinic or hospital setting capable of managing anaphylaxis.

Factors that may confer an increased risk for anaphylactic reactions to FIX include Hispanic ethnicity, a personal or family history of other allergies, and severe hemophilia B (FIX <1%) caused by total null mutations (both deletion and nonsense) in the F9 gene. It is unclear why anaphylactic reactions are more common with FIX deficiency than in FVIII deficiency. It is possible that the **extravascular distribution** of FIX is more likely to provoke such a reaction. In addition, therapeutic doses of FIX contain much more protein than therapeutic doses of FVIII; this may also contribute to the increased risk of anaphylaxis with FIX concentrates.

Whereas ITI is the preferred management for patients with hemophilia A and HTIs, a decision to attempt ITI for a patient with hemophilia B and HTIs must take into consideration the relatively high risk of severe complications (including **nephrotic syndrome** which is not always reversible with cessation of ITI, and anaphylaxis) and overall lower success rate (estimated at 30%). Highly **immunosuppressive** regimens, involving the combined use of rituximab, dexamethasone, or **mycophenolate mofetil** (MMF) with the ITI have shown better results...
in eradicating HTIs to FIX than ITI alone. Due to its rarity, little is known about the predictors of successful ITI in hemophilia B.

Although emicizumab, being a mimic for FVIII, cannot be used in patients with hemophilia B and inhibitors, other non-factor therapies such as fitusiran and anti-TFPI can. Further studies on this rare group of patients are needed.

The future of inhibitor management

Ongoing efforts continue on how best to prevent inhibitors, and for those patients that develop inhibitors on how best to eradicate these. We expect all of this to evolve considerably in the next months to years.

Manufacturers are working on developing new agents to treat and prevent bleeding in patients with inhibitors. Several companies are working on extending the half-life of rFVIIa either through PEGylation technology or by fusing rFVIIa to albumin. Extended half-life rFVIIa has the potential to significantly reduce the burden of treating and preventing bleeds.

As mentioned earlier, a number of FVIII substitutes have either been made available (e.g., emicizumab) or are in various stages of development. As the development of these novel agents is rapidly evolving, the reader is advised to consult the most recent literature for their current status.
Glossary

**albumin:** A protein found in human plasma that is used as a stabilizer in factor VIII and factor IX products including recombinant factor concentrates. Some new concentrates now use sucrose instead of albumin as a stabilizer.

**allo-antibody:** An antibody produced by the immune system in response to exposure to an antigen that is not present in the person’s own body, for example as a result of infusion of replacement factor concentrates. Allo-antibodies to factor VIII or IX that occur in people with hemophilia are called inhibitors.

**antibody:** Proteins made by the body’s immune system to fight off substances it perceives as foreign.

**auto-antibody:** An antibody produced by the immune system that targets antigens present in an individual’s own body, in contrast to allo-antibodies, which target antigens not originally present in the person’s own body. For example, acquired hemophilia A occurs when a person develops antibodies to their own factor VIII.

**anamnesis:** A quick rise in inhibitor titre following exposure to factor.

**anaphylaxis:** A severe allergic reaction often resulting in the inability to breathe.

**antifibrinolytic therapy:** A drug that can help stop the normal breakdown of blood clots and help speed recovery from a bleed. Also called fibrinolytic inhibitors.

**B-domain deleted rFVIII:** A concentrate of recombinant factor VIII that has had its B domain removed. This deletion increases the manufacturing yield of the product but does not impair *in vitro* or *in vivo* functionality of the recombinant factor.

**Bethesda assay:** A laboratory test to detect the presence of FVIII or FIX inhibitors in patient plasma.

**Bethesda Unit (BU):** A measurement of the level of inhibitors in blood, defined as the amount of inhibitor that neutralizes 50% of 1 unit of clotting factor during a given incubation period.

**buffer:** A chemical agent used in laboratory testing to maintain the acidity (pH) of a solution when mixed with other compounds. The original Bethesda assay to detect and quantify inhibitors set out the use of buffered pooled normal plasma in the control sample to correct for factor deterioration during incubation and improve FVIII/FIX stability and assay specificity; the Nijmegen modification of the Bethesda assay further standardizes the test and enhances assay reliability by buffering both the test and control dilutions with the addition of 0.1M imidazole to pH 7.4 along with use of immunodepleted FVIII/FIX-deficient control plasma.

**bypassing agent:** A special clotting factor used in patients with antibodies (inhibitors) to their usual factor, to overcome the blockage or cessation in the clotting system.

**clotting factor:** Any of the factors in blood plasma that work together to form a clot to help stop bleeding. Deficiency or absence of factor VIII (FVIII) clotting activity results in hemophilia A, while deficiency or absence of factor IX (FIX) clotting activity results in hemophilia B.

**consumptive coagulopathy:** A condition in which blood clots form throughout the body, blocking small blood vessels. Also referred to as disseminated intravascular coagulation.

**desmopressin (DDAVP):** A synthetic compound that raises a person’s factor VIII level in blood, but is not a blood product. It can be used to treat mild and, in some cases, moderate hemophilia A and some types of von Willebrand disease. It is administered intravenously, by subcutaneous injection, or by intranasal spray.

**dexamethasone:** A potent synthetic analogue of cortisol, with similar biological action; used as an anti-inflammatory agent.

**ELISA:** Enzyme-linked immunosorbent assay that detects and measures antibodies immunologically.

**emicizumab:** A recombinant humanized bispecific monoclonal antibody that bridges activated FIX and FX to mimic the function of missing activated FVIII in hemophilia A patients.
**epitope:** The simplest form of an antigenic determinant, on a complex antigenic molecule, which can combine with antibody or T cell receptor. The smallest part of a protein that an antibody recognizes.

**exposure day (ED):** An exposure day is a day on which a person with hemophilia has been infused with factor concentrate to treat or prevent a bleed. The number of EDs consists only of those days on which factor was infused.

**extended half-life (EHL) factor:** A new generation of recombinant factor concentrates based on strategies such as PEGylation, fusion technologies, and amino acid sequence modification designed with the intention of increasing the half-life.

**extravascular distribution:** The process by which a drug or protein passes from the bloodstream to body tissues and organs, from the intravascular space, e.g. blood vessels to extravascular spaces, e.g. body tissues, as it is carried around the body by the circulatory system.

**factor concentrate:** A type of hemophilia treatment that replaces the missing FVIII or FIX by injection into a vein. Factor concentrates can be manufactured from human plasma or by recombinant technology. They are purified and treated to destroy any potential viruses or diseases, then freeze-dried to a powder and stored in sterile vials. Before an infusion, sterile water is added to the clotting proteins for reconstitution.

**factor recovery:** The amount of infused factor concentrate that is actually utilized by the body to stop bleeding.

**fitusiran:** An investigational molecule for the treatment of hemophilia A or B patients with and without inhibitors that targets anti-thrombin to improve the coagulation equilibrium and promote sufficient thrombin generation to restore hemostasis and prevent bleeding.

**glycosylation:** Biochemical modification of a substance (usually a protein) by the addition of sugar molecules.

**half-life:** The time it takes for infused factor to lose half of its potency. Conventional FVIII has a half-life of 8 to 12 hours. Conventional FIX has a half-life of 18 to 24 hours. The half-life of EHL FVIII is approximately 1.5 fold longer than that of conventional FVIII, and that of EHL FIX approximately 3-5 fold longer than conventional FIX.

**haplotype:** A set of genetic determinants located on a single chromosome.

**historical peak inhibitor titre:** The highest inhibitor titre recorded in a patient before the start of immune tolerance induction therapy.

**imidazole:** A chemical agent used in laboratory testing to maintain the acidity (pH) of a solution when mixed with other compounds. In inhibitor testing, imidazole is used as a buffer to correct for factor deterioration during incubation and improve FVIII/FIX stability and assay reliability and specificity; the Nijmegen modification of the Bethesda assay stipulates buffering both the test and control dilutions with the addition of 0.1M imidazole to pH 7.4.

**immune depletion:** A method for removing a target molecule from a mixture.

**immune tolerance induction (ITI):** The infusion of high doses of the missing clotting factor concentrate 3-7 times per week for very long periods of time – months or years. The objective of the therapy is to allow the body’s defenses to become accustomed to the foreign factor and to stop making antibodies against it, so that normal doses will be effective in stopping bleeding.

**immunogenicity:** The ability of a particular substance, such as an antigen, to provoke an immune response.

**immunoglobulin (Ig):** Blood components responsible for immune function (defending the body against infection or playing a role in modulating the body’s immunological mechanisms). This component can be separated out during fractionation.
**immunoglobulin G (IgG):** The most abundant of the 5 classes of structurally related antibodies in the body. There are 4 subclasses of IgG (IgG1, IgG2, IgG3, and IgG4) antibody molecules. IgG is composed of four peptide chains: two heavy chains γ and two light chains. Each IgG has two antigen binding sites.

**immunosuppression:** Prevention or interference with the development of immunologic response.

**incidence:** The number of new cases of a disease in a population over a period of time.

**inhibitors:** Antibodies produced by the immune system against infused factor VIII or factor IX that attack and destroy the FVIII or FIX proteins in factor concentrates, making treatment ineffective.

**in vivo:** A process taking place in a living organism. This is in contrast to **ex vivo** – a process occurring outside the living organism.

**local hemostatic measures:** Measures to control bleeding that are applied locally, such as for dental surgery or postoperative bleeding.

**myocardial infarction:** A heart attack.

**mycophenolate mofetil (MMF):** An immunosuppressant drug used in combination with other medications to suppress the body’s immune system such as to help the body accept organ transplantation; in hemophilia, MMF is used in immune tolerance induction therapy to help eradicate inhibitors to factor concentrates.

**nephrotic syndrome:** A condition in which damage to the kidneys results in loss of proteins into the urine causing diffuse swelling (edema).

**null mutation:** A mutation in a gene that results in no protein (e.g., factor) being produced.

**pharmacokinetics:** The action of drugs in the body over a period of time, including the processes of absorption, distribution, localization in tissues, biotransformation, and excretion.

**plasma-derived factor concentrate:** Factor concentrates that have been fractionated from human blood. Plasma-derived concentrates are available that contain factor I (fibrinogen), factor VIII, factor IX, von Willebrand factor, factor XI, and factor XIII, or a mixture of factors II, VII, IX, and X (these are known as prothrombin complex concentrates).

**plasmapheresis:** A method of removing plasma from the body by withdrawing blood, separating it into plasma and cells, removing the plasma (which contains antibodies) and transfusing the cells back into the bloodstream.

**pooled normal plasma (PNP):** Plasma from a number of normal healthy blood donors is pooled together to obtain sufficient levels of factors and other blood components for fractionation. In the manufacturing of plasma-derived pharmaceutical drugs, the pooled plasma is subjected to rigorous viral testing and viral inactivation prior to fractionation into its component parts such as clotting factors, albumin and immunoglobulins.

**porcine FVIII:** FVIII concentrate made from the blood of pigs, mainly used to treat people with factor VIII inhibitors. Porcine rFVIII is a recombinant B-domain deleted form of FVIII that is typically not as quickly inactivated or destroyed as human FVIII is when administered to patients with inhibitors. It is available in some countries for the management of acquired hemophilia A, and may also become available for patients with hemophilia A and LTIs.

**prevalence:** The total number of cases of a disease in a given population at a specific time.

**previously treated patients (PTP):** People with hemophilia who have received at least 150 exposures to factor; sometimes this is defined as patients who have received at least 50 exposures to factor.

**previously untreated patients (PUP):** People with hemophilia who have not as yet received 50 exposures to factor and consequently are more vulnerable to inhibitor development.

**prognostic factors:** Characteristics that define the natural history of a disease, including predictive factors that tell whether a particular therapeutic intervention will result in a favorable outcome.
recombinant factor concentrate: A type of factor concentrate that is manufactured in a laboratory using recombinant (genetic) technology instead of being derived from human blood. Recombinant proteins are copies of certain kinds of proteins found in human blood plasma.

rituximab: A chimeric monoclonal antibody against the B-cell antigen CD20 (on B lymphocytes) that induces a rapid in vivo depletion of normal B lymphocytes. Primarily developed to treat B-cell non-Hodgkin lymphomas, rituximab has demonstrated effectiveness in a number of autoantibody-mediated diseases.

sulfation: Biochemical modification of a substance (usually a protein) by the addition of sulfur containing molecules.

thrombin generation assay (TGA): A test to detect the levels of thrombin generated in a patient. Determining the rate of thrombin generation can help indicate if patients are at risk of clotting or bleeding.

thromboelastography (TEG): A method of testing the efficiency of blood coagulation by measuring elastic variations of a thrombus (blood clot) during the coagulation process, mainly used in surgery and anesthesiology.

thrombosis: The formation of a blood clot within a blood vessel (artery or vein).

thrombotic microangiopathy (TMA): A pathology that results in thrombosis (formation of blood clots) in capillaries and arterioles, due to an endothelial injury.

titration: A laboratory method for determining the amount of a constituent in a solution by measuring the volume of a known concentration of reagent required to complete a reaction with it. In hemophilia, the Bethesda assay uses titration to determine the amount of inhibitors in a patient sample, referred to as inhibitor titre.

tolerization: A patient is “tolerized” when the inhibitor to FVIII or FIX has disappeared and does not re-appear with further treatment of FVIII or FIX.
### Acronyms and Abbreviations

<table>
<thead>
<tr>
<th>Acronym</th>
<th>Definition</th>
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<tr>
<td>aPCC</td>
<td>activated prothrombin complex concentrates</td>
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<td>BU</td>
<td>Bethesda Unit</td>
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<tr>
<td>BHK</td>
<td>baby hamster kidney cell line</td>
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<tr>
<td>CDC</td>
<td>Centers for Disease Control (United States)</td>
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<td>CHMP</td>
<td>Committee for Medicinal Products for Human Use (European Medicines Agency)</td>
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<td>CHO</td>
<td>Chinese hamster ovary cell line</td>
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<td>DDAVP</td>
<td>desmopressin</td>
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<td>ED</td>
<td>exposure day</td>
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<td>ELISA</td>
<td>enzyme-linked immunosorbent assay</td>
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<td>EMA</td>
<td>European Medicines Agency</td>
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<td>EHL</td>
<td>extended half-life</td>
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<tr>
<td>Fc</td>
<td>fragment crystallizable</td>
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<tr>
<td>FDA</td>
<td>Food and Drug Administration (United States)</td>
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<td>FEIBA®</td>
<td>Factor Eight Inhibitor Bypassing Activity</td>
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<tr>
<td>FII, FIIa</td>
<td>factor II, activated factor II</td>
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<td>FVII, FVIIa</td>
<td>factor VII, activated FVII</td>
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<td>FVIII</td>
<td>factor VIII</td>
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<td>FIX, FIXa</td>
<td>factor IX, activated factor IX</td>
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<tr>
<td>FX, FXa</td>
<td>factor X, activated factor X</td>
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<tr>
<td>HTI</td>
<td>high-titre inhibitor</td>
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<tr>
<td>IgE</td>
<td>Immunoglobulin E</td>
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<tr>
<td>IgG</td>
<td>Immunoglobulin G (IgG1, IgG2, IgG3, IgG4)</td>
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<tr>
<td>ITI</td>
<td>Immune tolerance induction (therapy)</td>
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<tr>
<td>IU</td>
<td>international unit</td>
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<tr>
<td>ISTH</td>
<td>International Society on Thrombosis and Haemostasis</td>
</tr>
<tr>
<td>LTI</td>
<td>low-titre inhibitor</td>
</tr>
<tr>
<td>MMF</td>
<td>mycophenolate mofetil</td>
</tr>
<tr>
<td>PEG</td>
<td>polyethylene glycol</td>
</tr>
<tr>
<td>PNP</td>
<td>pooled normal plasma</td>
</tr>
<tr>
<td>PTP</td>
<td>previously treated patient</td>
</tr>
<tr>
<td>PUP</td>
<td>previously untreated patient</td>
</tr>
<tr>
<td>PWH</td>
<td>people with hemophilia</td>
</tr>
<tr>
<td>rFVIIa</td>
<td>recombinant activated factor VII</td>
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<tr>
<td>rFVIII</td>
<td>recombinant factor VIII</td>
</tr>
<tr>
<td>RODIN</td>
<td>Research of Determinants of Inhibitor Development</td>
</tr>
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<td>Survey of Inhibitors in Plasma-Product Exposed Toddlers</td>
</tr>
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<td>TEG</td>
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<tr>
<td>TFPI</td>
<td>tissue factor pathway inhibitor</td>
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<td>TGA</td>
<td>thrombin generation assay</td>
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<tr>
<td>TMA</td>
<td>thrombotic microangiopathy</td>
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<tr>
<td>UDC</td>
<td>Universal Data Collection (U.S. Centers for Disease Control)</td>
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<tr>
<td>VWD</td>
<td>von Willebrand disease</td>
</tr>
<tr>
<td>VWF</td>
<td>von Willebrand factor</td>
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<td>WFH</td>
<td>World Federation of Hemophilia</td>
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References


