

TREATMENT OF HEMOPHILIA

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# MILD HEMOPHILIA

Revised edition

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**WFH**

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YEARS OF ADVANCING  
TREATMENT FOR ALL

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WORLD FEDERATION OF HEMOPHILIA  
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## Introduction

This monograph discusses the mild forms of hemophilia A and hemophilia B, from epidemiology and molecular basis, via diagnosis, to the treatment options available. The emphasis is on features that are characteristic of the mild form. Therefore, for example, treatment with factor concentrates will only be briefly mentioned.

## Classification

The severity of hemophilia has been defined by a traditional classification into three forms:

- Severe form: factor level  $<0.01$  IU/ml ( $<1\%$  of normal)
- Moderate form: factor level  $0.01$  to  $0.05$  IU/ml (1 to 5% of normal)
- Mild form: factor level  $>0.05$  to  $0.40$  IU/ml (more than 5 to 40% of normal)

This definition has been published by the subcommittee on Factor VIII and Factor IX of the Scientific and Standardization Committee of the International Society on Thrombosis and Haemostasis [1].

However, particularly the upper limit for mild hemophilia is vague and may in different publications vary from  $0.25$  IU/ml (25%) up to  $0.50$  IU/ml (50%), which is the lower limit of the normal range. The wider the range for the definition of mild hemophilia, the greater the proportion of females who fall into it will be. For example, in one study, using the definition 5 to 50% of normal factor levels, the proportion of females was 10%[2]. Females, who are generally carriers of hemophilia, have the same risk of bleeding as a male with mild hemophilia at the corresponding factor level. Carriers report significantly more bleeding events than non-carriers from small wounds and after invasive procedures, and their bleeding tendency is inversely correlated to their factor level [3].

## Epidemiology

The prevalence of hemophilia in countries where diagnostic tools are readily available is about 1 in 10,000 people. The proportion of those with a mild form of hemophilia varies between countries, over time in the same country, and between the two types of hemophilia (A and B). This variation to a considerable extent depends on the resources available and the awareness of hemophilia among physicians. In the World Federation of Hemophilia's *Annual Global Survey 2004*, the proportion of patients diagnosed with mild hemophilia (34%) was close to the proportion with severe hemophilia (43%) in countries with a gross natural product (GNP) per capita of more than US\$10,000. In countries with a GNP below US\$2,000 per capita the proportion with mild hemophilia (18%) was clearly lower than that of the severe form (50%).

In a Swedish survey of all patients with hemophilia, the proportion of confirmed cases of mild hemophilia increased from 35% in 1960 to 54% in 1980[4]. In this case the change is due to increased awareness and family investigations rather than to a change in GNP. In a survey of 147 hemophilia treatment centres worldwide, the prevalence of the mild form among patients with hemophilia A was 32%[5]. In small countries, the best example probably being Iceland, the distribution can be extremely skewed[6].

## Life expectancy

The course of the disorder is obviously less dramatic in mild compared to severe hemophilia, and the life expectancy is thus very close to that of the normal population. Whereas an impressive improvement in life expectancy was described in Swedish patients with severe hemophilia in the period from 1831 to 1920 (11 years) to the period from 1960 to 1980 (57 years), no such comparison could be made for mild hemophilia, due to lack of data from the early period. However, during the period of

1960–1980 the life-expectancy was 72 years for patients with mild hemophilia, compared to 75.5 years in the normal population [7].

This positive picture was tragically changed by the emergence of transfusion-transmitted viral infections, which affected patients with all forms of hemophilia. In the United Kingdom the death rate during the period of 1977 to 1984 of 4 per 1,000 with mild or moderate hemophilia increased to 85 per 1,000 in HIV-seropositive patients in 1991–1992 [8]. The general improvement of life expectancy for patients with mild hemophilia during the past century, derived from studies in Sweden [7], the United Kingdom [8], and the Netherlands [9, 10], is depicted in Fig. 1.

The leading causes of death during the past two decades in patients with mild hemophilia are infections with hepatitis C and with human immunodeficiency virus (HIV), similarly to the patients with severe hemophilia [10–13]. On the other hand, hemophilia provides partial protection against death from cardiovascular causes [10], although in one study the prevalence of ischemic heart disease was higher in mild hemophilia (3.4%) than in moderate (0.7%) or severe hemophilia (0.4%) [14].

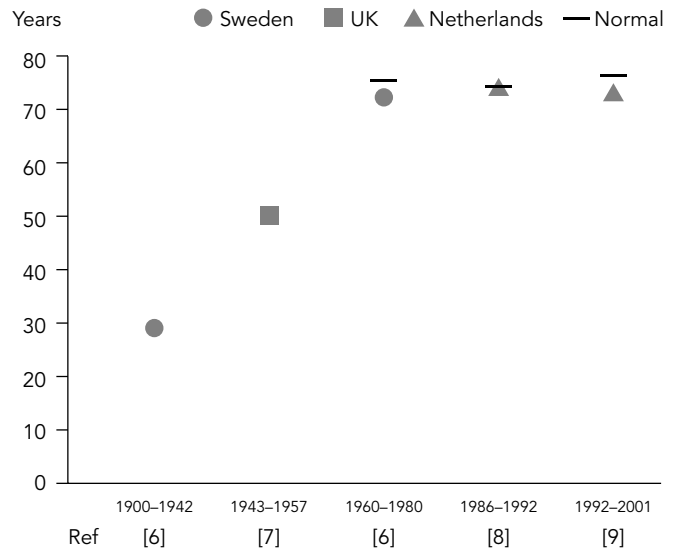
## Diagnosis

The diagnosis of mild hemophilia is often made as part of a family investigation, as was the case for 64% of patients diagnosed at two centres in the United States, for example [2]. In the other cases in this study the diagnosis was made after one or several bleeding episodes and at a mean age of 5.5 years. Among the latter, the presenting bleeding episodes were hematemesis, soft tissue or joint bleeding, or prolonged bleeding after surgery in the mouth or nose.

In a recent French study the diagnosis of mild hemophilia was already made at the age of 2.4 years [15]. However, from time to time elderly persons will be diagnosed with mild hemophilia as a result of an investigation triggered by bleeding complications after surgery or tooth extraction.

Bleeding is rarely spontaneous in patients with mild hemophilia. In fact, in the above-mentioned US investigation, 92% of the bleedings were precipitated by trauma [2]. The type of bleeding was less often in the joint (30%) than in soft tissues (53%).

**FIGURE 1.** Improvement in life expectancy of people with hemophilia (1900–2001)



## Laboratory diagnosis

Patients investigated for bleeding diathesis are typically first screened with analysis for platelet count, activated partial thromboplastin time (aPTT), prothrombin time, bleeding time, and/or platelet function analyzer (PFA)-100 test. Only the aPTT may be abnormally prolonged in patients with mild hemophilia, but this depends on the sensitivity of the reagent and on the deficient factor level [16]. Standardization of the method is crucial to obtain reliable and reproducible results [17]. Many hemophilia treatment centre laboratories missed the diagnosis of mild hemophilia using aPTT [18].

A factor assay must therefore be performed if there is clinical suspicion, even in the absence of a prolonged aPTT. Unfortunately, even with a routine factor VIII (FVIII) assay, the resulting clinical diagnosis can vary. In a study in the United Kingdom with plasma samples from three untreated patients with hemophilia distributed to a large number of centres, one sample with a median level of 5.8% yielded a range from 1.5% to 36% [19]. The most accurate results were obtained from comprehensive care centres. It is thus important for every centre to participate in a quality assurance program for the assays of FVIII and factor IX (FIX), and also to react and improve the performance if the result is far from the expected number. The WFH plays an active part in this respect with support for centres participating in the WFH-sponsored International External Quality Assessment Scheme (IEQAS).

TABLE 1. Characteristics of mild hemophilia vs. VWD type 2N

	Mild hemophilia A	von Willebrand disease type 2N
<b>Inheritance</b>	X-linked	Autosomal recessive
<b>Mutation</b>	FVIII gene	VWF gene*
<b>Capacity of VWF to bind FVIII</b>	Normal	Reduced
<b>Response to DDAVP</b>	Good	Shorter effect
<b>Response to FVIII concentrate</b>	Good	Good only if VWF present in concentrate

\* The mutation in the VWF gene responsible for VWD type 2N results in amino acid substitution in the N-terminal part of the VWF molecule, where the FVIII binding site is located.

There are additional difficulties in the diagnosis of mild hemophilia using a factor assay in the case of certain molecular defects, as described below (see ‘Molecular basis for mild hemophilia’ on page 6). Furthermore, acute phase reaction after surgery or in inflammatory disease will temporarily increase the FVIII level, which also increases with age but is lower in people with blood group O [20, 21].

### Differential diagnosis

Mild hemophilia A has to be differentiated from the mild form of von Willebrand disease (VWD). The latter is often also characterized by a reduced FVIII level, but in addition there may be a prolonged bleeding time, a prolonged closure time in the PFA-100 instrument, and of course a reduction of the von Willebrand factor (VWF) level in plasma, which can be measured as antigen, ristocetin co-factor, collagen binding activity, FVIII binding capacity, multimer formation etc. The most challenging differential diagnosis is versus the von Willebrand Normandie variant (VWD type 2N) [22]. As in mild hemophilia A, the FVIII level is typically between 5% and 30% without a prolonged bleeding time or reduced plasma VWF level and the clinical picture may be similar. The differences between the two entities are shown in Table 1.

Obviously, a similar problem can occur if the defect is at the other side of the binding, i.e. on the FVIII molecule at its VWF-binding site in the C1-domain. Table 2 shows such variants according to the mutation and the resulting effect on the binding to VWF. These patients also have reduced secretion of functional FVIII and by definition they have hemophilia A.

Another differential diagnosis is combined deficiency of factor V (FV) and FVIII. Again, the FVIII levels are as in mild hemophilia A, but the inheritance of the combined deficiency is autosomal. The defect is neither in the F5 gene nor in the F8 gene, but in one of the “chaperone” proteins required for the post-translational processing and cellular secretion of these two structurally similar coagulation factors. The diagnosis is evidently verified by also measuring the FV level. Typically, both the FV and FVIII levels are in the 5 to 50% range.

Mild hemophilia B must be differentiated from conditions characterized by combined deficiency of the vitamin K dependent factors (in addition to FIX also factors VII, X, and prothrombin [FII]). This can be acquired due to vitamin K deficiency, liver disease, or the use of vitamin K antagonist drugs such as warfarin or it can be congenital due to a mutation in the gene for  $\gamma$ -glutamyl-carboxylase or for vitamin K epoxide reductase.

Severe hemophilia is sometimes associated with a mild phenotype, more commonly seen in hemophilia B than in hemophilia A, and the genetic basis is non-null mutations [23].

TABLE 2. FVIII gene mutations and effects on binding to VWF

Mutation in FVIII gene	Reduction of affinity to VWF
Ile2098Ser	8-fold
Ser2119Tyr	80-fold
Arg2150His	3-fold

## Molecular basis for mild hemophilia

### Hemophilia A

In a study of 101 patients with mild or moderate hemophilia A, most of whom were unrelated, Schwaab et al described that underlying missense mutations accounted for 86% of the patients [24]. These mutations may result in reduced synthesis, processing, secretion, or stability of FVIII, impaired thrombin activation, disturbed interaction with VWF or FIX, or decreased binding to phospholipids.

A minority of patients has a much more pronounced defect in FVIII activity than the amount of circulating

FVIII antigen, which is called Cross Reacting Material (CRM) positive. One explanation for this was found in patients with mutations causing a change in the electrical charge of the A2 domain of the FVIII molecule. This was responsible for a more rapid degradation of active FVIII [25], so although normal amounts are produced and secreted, the activity diminishes faster during the assay. This mutation also appears to be associated with discrepancies in the one- and two-stage clotting assays used for measuring FVIII.

The reverse phenomenon with higher two-stage clotting levels has been observed with the Tyr346Cys mutation,

**TABLE 3.** F8 mutations and the effect on clotting factor assays

Mutation	FVIII domain	One-stage clot	Two-stage clot	Function
<i>High ratio one-stage/two-stage</i>				
Pro264Leu [26]	A1	14–30%	5–16%	↑dissociation of A2
His281Asn [26]	A1	38%	25%	Not described
Ala284Glu [27,28]	A1	38%	10%	↑dissociation of A2
Ser289Leu [27,28]	A1	33%	9%	↑dissociation of A2
Arg527Trp [29]	A2	27%	13%	Not described
Arg531His [25,30]	A2	36%	19%	↑dissociation of A2
Arg531Cys [31]	A2	8–20%	3–8%	↑dissociation of A2
Asn694Ile [30,32]	A2	5–30%	2–10%	↑dissociation of A2
Arg698Leu/Trp [27,30]	A2	28–35%	6–15%	↑dissociation of A2
Arg1749His [26,33]	A3	71%	32%	Not described
Phe1785Leu [26]	A3	21%	13%	Not described
Ser1791Pro [29]	A3	19–32%	5–9%	Not described
Leu1932Phe [27]	A3	19%	7%	Not described
His1954Leu [34]	A3	106%	18%	↑dissociation of A2
Leu1978Phe [29]	A3	10%	2–4%	Not described
<i>Low ratio one-stage/two-stage</i>				
Tyr346Cys [35]	a1	34%	110%	Complex effects
Ile369Thr [26]	a1	9–20%	74–105%	Delayed activation
Glu720Lys [26]	a2	30–39%	99–115%	Not described
Arg1689His [29]	A3	25%	99–111%	Not described*
Phe2127Ser [26]	C1	3–25%	18–124%	Not described

\* Almost no bleeding in 4 patients

probably related to more efficient FVIII activation with the longer incubation time with thrombin in the two-stage assay. A few identified mutations and the effect on the assays are listed in Table 3. Generally, the mutations with higher levels measured in the two-stage than the one-stage assay appear to have an extremely mild phenotype, almost defining the low one-stage results as laboratory errors.

For some patients with no detectable mutation using DNA sequencing, deep intronic variations have been identified [36]. These may cause hemophilia by generation of new splice sites with premature termination codons. Rarely, mutations causing mild hemophilia A have been identified in the F8 promoter region [37].

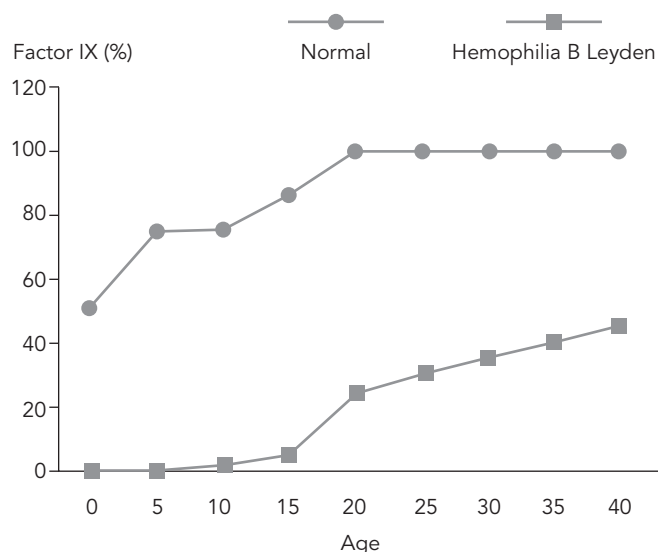
In certain geographical regions one mutation can dominate among patients with mild hemophilia due to a common founder effect. This has been described in Iceland [6]. Another example is Northern Italy, where 32% of the patients have a duplication of exon 13 in the F8 gene, which results in no activity of the molecule and should manifest as severe hemophilia. However, due to a phenomenon called “exon skipping” one of the exon 13 twins is sometimes not read (alternative splicing), resulting in a few normal FVIII molecules and thus a phenotype of mild hemophilia [38].

### Hemophilia B

It has been estimated that in 97% of patients with mild hemophilia B the underlying defect is a missense mutation [39]. The mutations can cause reduced interaction with factor VII-tissue factor and therefore reduced activation of FIX [40], decreased activity due to reduced affinity to FVIII [41], or reduced activity if the amino acid substitution is in the catalytic domain [42], which is a common mutation in the Amish population. Mutations in the carboxyterminal portion of FIX (residues 403-415) result in impaired secretion of the factor from the hepatocyte [43], but the secreted molecules have normal function.

The most spectacular type of mutation in the F9 gene results in low FIX levels until puberty, and thereafter a rise of 5% per year up to a maximum of about 60%. This was first described in 1982 by a Dutch group and is called hemophilia B Leyden [44]. It should be noted that even in the normal child, there is an increase of the FIX activity in parallel with the maturation of the liver, but here the

**FIGURE 2.** Increase in FIX levels with age in the normal population and in patients with hemophilia B Leyden



baseline is about 50% and a major rise occurs already during the first 5 years of life, with a second rise during puberty (Fig 2).

The point mutations associated with hemophilia B Leyden have been identified in the promoter region at nucleotide -20, -6, and -5, and in exon 1 at nucleotide +8 and +13. The promoter region of F9 contains a binding site for hepatocyte nuclear factor 4 (HNF-4), a transcription factor in the steroid hormone receptor superfamily [45]. In hemophilia B Leyden the binding of HNF-4 to this promoter region is impaired and thereby also the transactivation of FIX. This is partly overcome by the increasing level of testosterone in puberty.

### Treatment

The use of factor concentrates is vital in patients with mild hemophilia in case of major surgery or trauma. The principles are the same as for patients with severe hemophilia with identical target levels and need for frequent bolus doses, at least initially. The dose required to reach this target level, whether given as bolus doses or as continuous infusion, obviously decreases with increasing baseline level of the deficient factor. However, for patients with mild hemophilia there are additional treatment options.

For minor procedures in patients with factor levels in the upper range of mild hemophilia (approximately 20% factor level), treatment with an inhibitor of fibrinolysis may be sufficient. This approach, for example with tranexamic acid (20 mg/kg orally or 10 mg/kg intravenously every 8 hours), should be used for any surgery involving mucosal membranes. For dental extractions mouth rinse (“swish and swallow”) with tranexamic acid is efficient and safe (see WFH Treatment of Hemophilia Monograph No.40, *Guidelines for dental treatment of patients with inherited bleeding disorders*). Inhibitors of fibrinolysis should not be used for hematuria due to the risk for clots in the ureter with obstruction.

### Hemophilia A

In mild hemophilia A, an important alternative to factor concentrates is desmopressin (DDAVP). Thirty years ago Dr. Pier Mannucci discovered that DDAVP elevates the FVIII level about three-fold over baseline and is a useful strategy to provide hemostasis during surgical procedures in these patients [46]. The consequent and consistent use of this alternative mode of treatment saved many Italian patients with mild hemophilia A from transfusion-transmitted viral infections in the late 1970s and early 1980s.

A FVIII level of at least 30% should be reached after the infusion or injection to suffice for treatment of minor events but for major surgery the level should be above 50%. The dose (0.3 µg/kg) is preferably given subcutaneously 1 hour before the procedure and may have to be repeated every 8 to 24 hours, depending on the extent of the surgery. However, repeated doses may cause fluid retention with hyponatremia and seizures in sensitive subjects (such as children and women at the time of delivery). The effect of desmopressin may also decrease after several doses—a phenomenon known as tachyphylaxis.

The half-life of the released FVIII is 5-8 hours. Since the rise of FVIII is quite variable between patients (but reproducible from time to time), a test should be done in each patient to evaluate the response, most suitably in association with diagnosis. In a population of 62 patients with mild hemophilia, 47% responded to DDAVP with a doubling of the factor VIII level and a peak level above 30% [47]. Predictors for a good response were a higher baseline level and older age. In another study with 74 patients, the mutation in the F8 gene was also identified

as a predictor [48]. Specifically, patients with mild hemophilia without detectable F8 mutations have a poor response to DDAVP [49], as well as patients with the -257T>G mutation in the F8 promoter region [37].

Intranasal administration is also possible but requires higher doses.

DDAVP may be useful in patients with mild hemophilia A and an inhibitor [50], since the antibodies may not inhibit the endogenous FVIII or not to the same extent as exogenous FVIII.

Other treatment options are activated prothrombin complex concentrate or recombinant factor VIIa. For eradication of an inhibitor, rituximab is significantly more effective in patients with mild hemophilia compared to severe hemophilia, with a 75% versus 43% success rate [51].

### Hemophilia B

A small but certain benefit of DDAVP has also been reported in patients with mild hemophilia B. Although the FIX level did not increase much (1.4 times), there was a shortening of the aPTT and dental surgery could be performed with good result [52]. This is probably a result of compensation from high levels of FVIII and VWF. It should be emphasized that this effect of DDAVP is by no means as well documented as that in hemophilia A and it is of a much smaller magnitude in hemophilia B.

In patients with hemophilia B Leyden attempts have been made to antecede the effect of puberty by using anabolic steroids [53], but this is not a recommendable alternative due to many side-effects.

### Follow-up

Patients with mild hemophilia should not be left without follow-up after diagnosis. Although the intervals between visits can be longer than for those with severe hemophilia, a review of the patient every two to three years is vital. At this visit a bleeding history is taken, physical exam (especially to identify arthropathy) is performed, laboratory tests to identify complications of previous transfusions (viral markers, factor inhibitor) are done, and information is given to the patient and/or his/her parents. This



information should include signs and symptoms typical for routine and dangerous types of hemorrhage, as well as appropriate response and treatment intervention. Self-injection of desmopressin is useful to teach patients with mild hemophilia A who bleed at least once per year.

## Inhibitors

Inhibitors occur also in mild hemophilia, most typically after intensive exposure to factor concentrates [54]. Suspicions have been raised that continuous infusion of factor concentrates may be more risky in this respect than bolus injections [54,55].

The cumulative incidence of inhibitors in patients with mild hemophilia A has been reported to be 3 to 13% by the age of 33 in some populations. These patients often have missense mutations with conformational changes at the site of an antigenic epitope on the surface of the molecule. The most common mutations that generate an inhibitor are Arg593Cys and Trp2229Cys. Patients with these mutations have a 40% risk of developing an inhibitor [56]. In 60% of these patients, tolerance occurs spontaneously about nine months after appearance of the inhibitor, but in the meantime the inhibitor can cause severe bleeding and even death. Inhibitors in patients with mild hemophilia B are extremely uncommon.

## Quality of life

Patients with mild hemophilia report better health-related quality of life (HR-QoL) than those with severe hemophilia [57,58], yet in several studies the levels were lower than in the normal population [58-60]. This is only partly explained by transfusion-transmitted viral infections


[59]. A more important contributor seems to be joint damage as a result of prior hemarthroses [60], which stresses the importance of early diagnosis and appropriate management.

## Conclusion

Mild hemophilia is a neglected diagnosis. Patients with mild forms of hemophilia have not had the same degree of medical attention as those with severe forms. Although justified to a certain extent, this has occasionally resulted in serious neglect. One of the first reports was in 1964. Due to a delayed diagnosis, an 18-year-old male with trauma-induced hemarthrosis that eventually resulted in infection, osteomyelitis, and life-threatening sepsis, required amputation of the leg at the hip joint [61].

A review of all Swedish patients with hemophilia in 1982 demonstrated the ironic situation that the mortality in bleeding in the central nervous system was higher in mild than in severe hemophilia [62]. More recently, in a Belgian-French study, intracranial hemorrhages were identified in 83 patients with severe hemophilia with fatal outcome in 17 (20%), and in 40 patients with mild hemophilia, with fatal outcome in 10 (25%).

Carriers of hemophilia with factor levels in the range of mild hemophilia should receive the diagnosis of mild hemophilia, since there is a widespread concept that males have the disease and females are “only” carriers.

It is of utmost importance to disseminate knowledge about mild hemophilia, to keep patients informed and updated by regular visits, and to implement appropriate diagnostic tools and treatment modalities. 

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