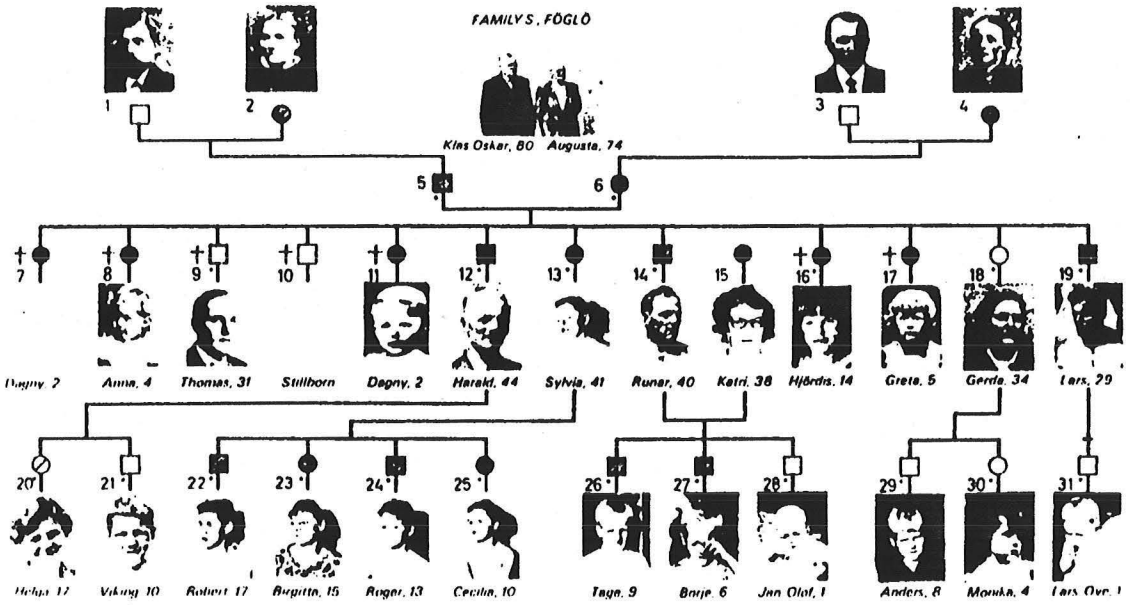
Von Willebrand's Disease
Diagnosis and Management



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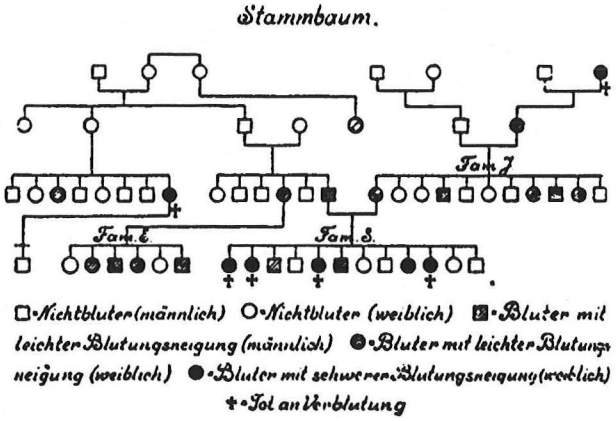
Her main clinical interests are in "benign" hematologic disorders, including sickle cell disease and disorders of hemostasis. Her current research centers around new immunological approaches to treating immune thrombocytopenic purpura.

HISTORICAL BACKGROUND

Von Willebrand's disease is the most common inherited bleeding disorder, estimated to occur in approximately one per cent of the population, and up to two per cent in Scandinavian countries. Its name honors Erik von Willebrand, who described the first kindred with this disorder in 1926 [1-3]. Von Willebrand was a Finnish internist who had first learned of this familial bleeding disorder when, as a young man, he had worked as a spa physician in the Åland Islands in the Gulf of Bothnia, between Sweden and Finland in the late nineteenth century. He subsequently became a medical academic in Helsinki, studying and writing in diverse medical fields, including iron deficiency, diabetes and obesity. In 1925 he was asked to see a 5 year old girl brought by her desperate parents from the Åland Islands. She suffered from a pronounced bleeding tendency - she was subsequently to die from menorrhagia at the age of 14 - and three of her sisters had died of bleeding between the ages of two and four. Both her parents had mild bleeding histories, as did many individuals in several generations on both sides of the extended family[4].

Intrigued, von Willebrand went back to the Åland Islands and studied the kindred with the simple coagulation studies then available, namely the platelet count, bleeding time, and the whole blood clotting time. Twenty three of the 66 family members he studied had a significant bleeding history. He found affected individuals had a pattern of prolonged bleeding times, despite normal platelet counts, whereas clotting times were normal.

Von Willebrand found considerable variation in the clinical severity of the disease and a pattern of transmission that suggested autosomal dominant inheritance. The only severely affected individuals were the index case and her siblings.

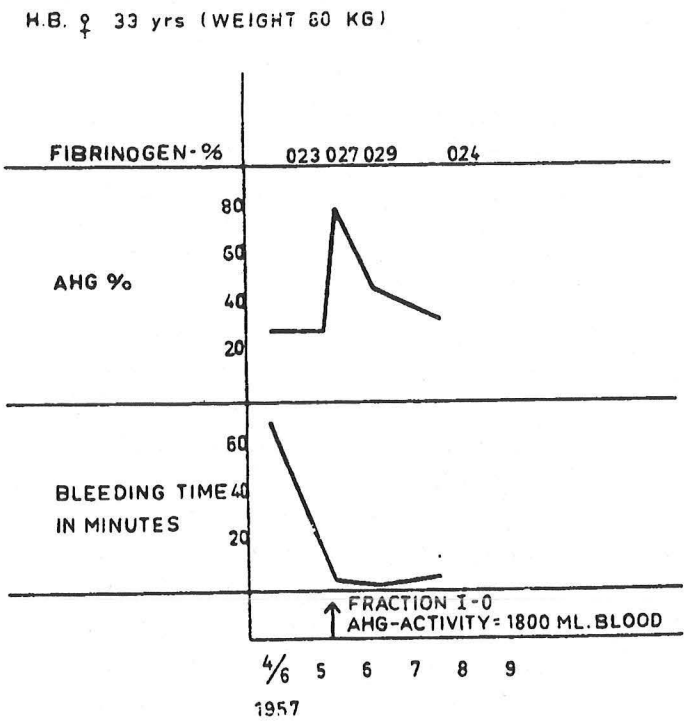


Family tree of von Willebrand's original kindred

Von Willebrand recognized that this was a new syndrome. The dominant pattern of inheritance distinguished it from the inherited platelet function disorder, Glanzmann's thromobasthenia. Both the occurrence of female cases, and the prolonged bleeding times distinguished this disorder from classical hemophilia, so he called this disorder "hereditary pseudohemophilia", and believed there were both vascular and platelet defects.

In the early 1950's methods were developed for measuring factor VIII levels (then called anti-hemophiliac globulin, or AHG). Several series of patients with hemorrhagic syndromes were described in which there was both a prolonged bleeding time and a reduced factor VIII level, including a series described in Sweden by Inga Marie Nilsson, a venerated and still active researcher in this area [5]. She subsequently took her coagulation laboratory to the Åland Islands, linked up with von Willebrand's original family, including the next generation, and demonstrated that they also had this pattern of deficient AHG and prolonged bleeding time [6].

Nilsson and her colleagues were able to extend the studies dramatically further, both in elucidating the disease and preparing the ground for specific treatment. Cohn fractionation of plasma was known to yield a product, fraction I-O which was rich in fibrinogen and anti hemophiliac globulin. They showed that infusing the I-O fraction into a patient with von Willebrand's disease both restored the factor VIII level and normalized the bleeding time [7]. They noticed that the factor VIII level stayed elevated much longer than when the I-O fraction was infused into hemophiliacs, and even found the I-O fraction from hemophiliac (factor VIII deficient) plasma would restore the VIII level in vWD and normalize the bleeding time. This was strong evidence that the deficient component in von Willebrand's disease was different from, though clearly closely linked to factor VIII [8].



Administration of AHG to a patient with Von Willebrand's disease

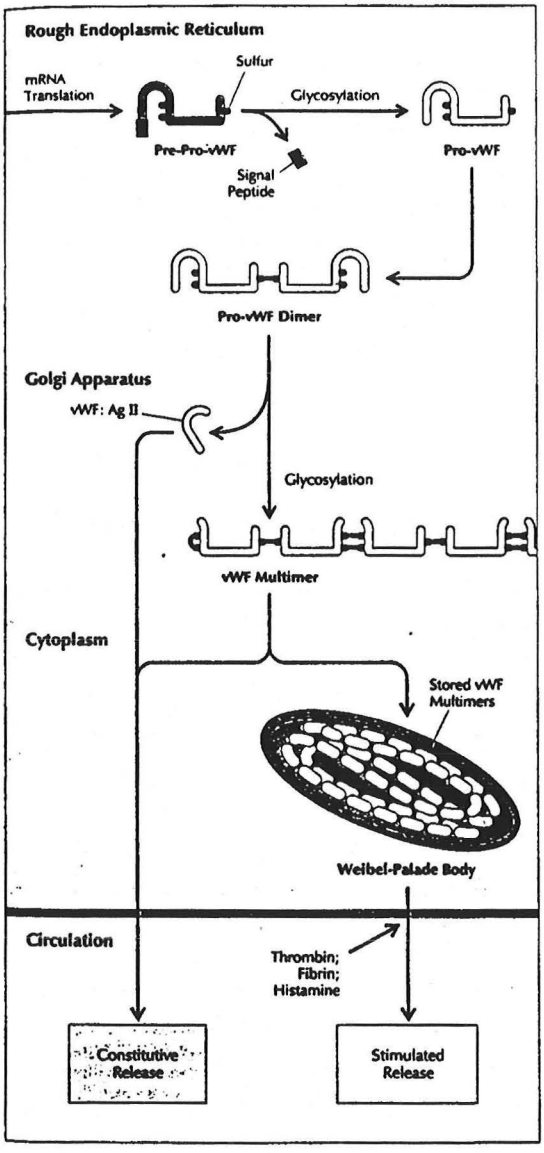
In 1985 the cDNA for von Willebrand's factor was successfully cloned, a breakthrough in understanding the role of von Willebrand factor in primary and secondary hemostasis. Specific functional domains on the von Willebrand factor molecule have been identified by studying different types of mutations, in the context of their clinical and laboratory phenotypes [9-12]. Binding sites for platelet receptors, collagen and factor VIII have been identified [13]. Over the past decade numerous mutations and polymorphisms have been described. A database of these is maintained on the Internet at http://mmg2.im.med.umich.edu[14]. Grouping the mutations into functional classes has enabled a simplified functional classification of vWD [15].

BIOSYNTHESIS OF VON WILLEBRAND'S FACTOR

Von Willebrand's Disease is an inherited disorder associated with either quantitative or qualitative deficiency of von Willebrand's factor (vWF), a multimeric glycoprotein, the synthesis of which is controlled by a gene on autosomal chromosome 12. The vWF is a high molecular weight glycoprotein synthesized by both endothelial cells and megakaryocytes.

The initial protein derived from the vWF gene has 2813 amino acid residues and is called pre-pro-vWF. The signal peptide which is 22 amino acids long is cleaved off in the endoplasmic reticulum, and the remaining protein undergoes glycosylation, creating the pro-vWF monomer. These monomers rapidly dimerize through disulfide bonds and are transported to the Golgi apparatus where the 763 amino acid pre-sequence is cleaved off. The remaining dimers are linked head-to-tail by further disulfide bonds, to assemble a whole series of multimeric proteins of varying molecular weights. The pro-sequence is clearly important in this process and in storage of the vWF. Without it transfection experiments showed that vWF monomer can be synthesized, but cannot form dimers or multimers [16].

The vWF multimers are secreted constitutively by endothelial cells; this secretion is the main source of von Willebrand's factor in plasma which circulates at about 7-10 µgram per mL. This plasma vWF includes a range of multimers from 500 to 20,000 kilodaltons, making it the largest protein in human plasma.



vWF secretion in endothelial cells [16]

Some of the largest vWF multimers are up to 2μm in length, comparable to normal platelet diameter. Certain physiologic and pharmacologic stimuli can cause endothelial cells to release vWF; this involves the release of multimers which have been previously synthesized and stored in special intracellular organelles called Weibel-Palade bodies. This stored component is very rich in high molecular weight multimers, including some even larger than those found in plasma, referred to as "unusually large multimers".

vWF is also synthesized by megakaryocytes in a similar way, except that formation of multimers is more complete. These multimers are stored in the alpha granules of megakaryocytes and their platelet offspring, which contain a higher concentration of the very high molecular weight forms of vWF [17]. This vWF is only released when the platelets are activated, after which the von Willebrand factor is found in very high concentration on the platelet membrane.

The vWF multimers are composed of 270-kD polypeptide subunits, each of which have binding sites for factor VIII, for collagen and for platelet glycoproteins GPIB and GPIIb/IIIa.

THE FUNCTION OF VON WILLEBRAND'S FACTOR IN HEMOSTASIS

Von Willebrand's factor has two main functions, both of which are fundamental to hemostasis:

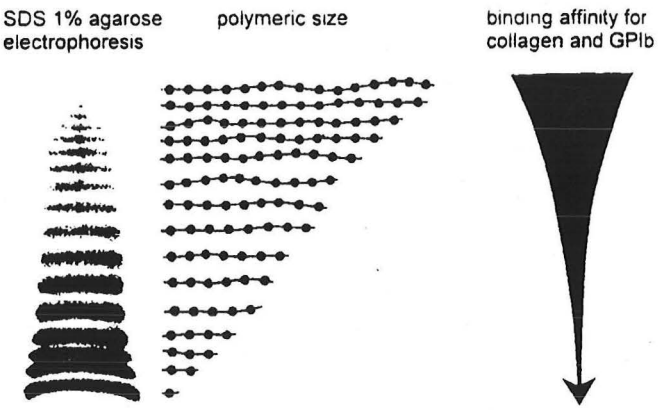
1. It acts as the **transport protein for coagulant factor VIII**, delivering it to the sites of vascular injury, where it participates in the intrinsic coagulation cascade. vWF and factor VIIIc circulate in plasma as a non-covalently bound complex, consisting of about one percent factor VIII and 99 percent vWF. This linkage protects factor VIII from inactivation by activated protein C and activated factor X. If the vWF level is reduced, or there is an abnormality in the interaction between vWF and factor VIII, factor VIII has a shortened half life [13, 18].

2. In primary hemostasis **vWF acts as a ligand between platelet glycoprotein receptors and exposed vascular sub-endothelium** at sites of injury. This involves a reactions between vWF and two different platelet receptor glycoproteins, GPIb/IX and GP IIb/IIIa and with components in the sub-endothelium. Although the GPIb receptor is exposed on the platelet membrane, under normal blood flow conditions the vWF circulating in the plasma does not bind to this receptor. However, under conditions of increased shear stress, such as occur at sites of injury, the vWF will bind to the receptor, which then activates the platelet causing it to express GPIIb/IIIa on its surface. This activated IIb/IIIa then can bind to many substances, including vWF, fibrinogen, fibronectin and vitronectin, forming cross-links between platelets, i.e. platelet aggregation [11, 19, 20].

A number of non-physiological stimuli can also induce the exposure of the GPIb receptor and initiate platelet aggregation: these include the antibiotic ristocetin (which was withdrawn from clinical use because it caused platelet aggregation and thrombocytopenia) [21, 22]. and the snake venom, Botrocetin; removing the negatively charged sialic acid residues from vWF carbohydrate side chains also causes the GPIb receptor to be exposed. Platelet activation then causes secretion of platelet vWF, which is expressed on the surface membrane, and which binds to collagen and other components of the subendothelium.

SDS get electrophoresis shows that both plasma and platelet vWF contain a variety of multimers; their regular spacing on the gel shows that each differs by a constant factor of approximately 500kD from adjacent bands[23]. This distribution of multimers is believed to be due to their ongoing proteolytic degradation.

Only the larger multimers appear to function in hemostasis [9, 24, 25]. When vWF functional studies are performed with multimers eluted from gels, the function drops off as the as the fragments become smaller. Similarly there is a loss of function when vWF multimers undergo disulfide reduction - the smaller the fraction the less hemostatically active.



Size and function of vWF multimers [9]

THE VON WILLEBRAND FACTOR MOLECULE

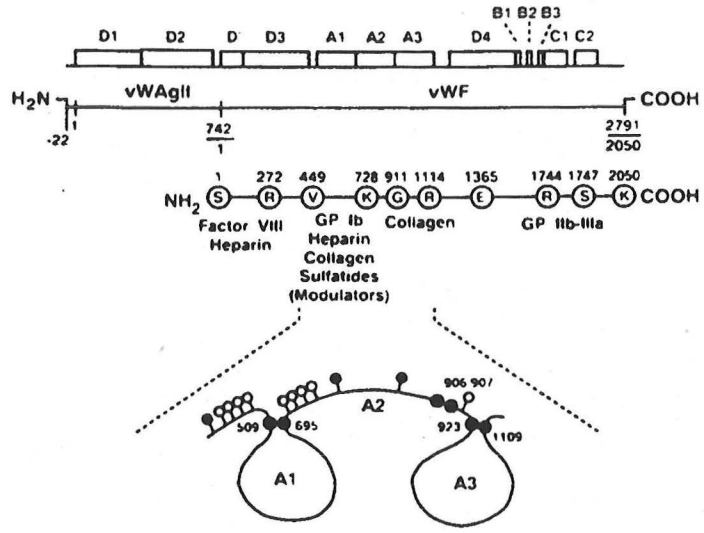
The vWF molecule (monomer) contains a number of functional domains, which include the structural components for its multiple functions and interactions:

The binding site for factor VIII is believed to lie within the first 272 amino acids, probably between residues 78-96, as ascertained through study of amino acid substitutions in these areas which diminish factor VIII binding.

The three A domains of the molecule contain a structural motif common to many adhesive proteins; these are known to be the domains in vWF involved in binding to ligands such as GPIb, collagen, etc. The A3 domain and to a lesser extent the A1 domain interact with collagen types I and III;

domain A1 binds to collagen type VI and also contains a site for binding to heparin. A1 is also believed to contain the vitally-important binding site for platelet GPIb, which is usually not exposed until the molecule undergoes a conformational change upon binding to the subendothelium [20, 25].

The binding of vWF to GPIIb/IIIa appears to be exclusively mediated through a domain near the carboxyl-terminal region



Structure of the Pre-pro vWF and the functional domains in mature vWF [25]

CLASSIFICATION OF VON WILLEBRAND'S DISEASE

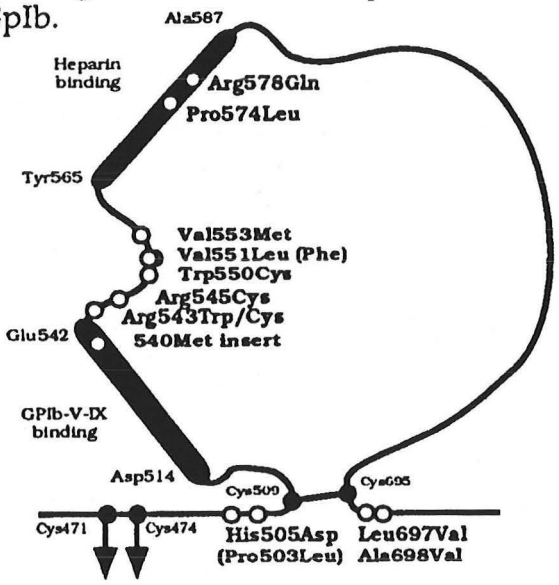
Classification of Von Willebrand's disease has recently been simplified, to a system according to the type of the molecular defect, and the laboratory testing, especially the pattern of the vWF multimers. (Note use of Arabic instead of Roman numerals). This classification correlates well with the clinical picture and the treatment required [15].

Type 1 is a quantitative deficiency in vWF. This is by far the most common type and constitutes 70 to 80 percent of all cases. It is inherited in an autosomal dominant manner. The molecular pathogenesis of type 1 von Willebrand's disease is unclear. No mutations of the vWF gene have been associated with this subtype. It is possible that there will be more than one molecular mechanism identified for this common form of von Willebrand's disease.

Type 2 von Willebrand's disease includes patients with a variety of **qualitative** defects in vWF. Inheritance is usually autosomal dominant. This type constitutes 15 to 20 percent of cases. Type 2 is further subdivided into:

Type 2A: qualitative variants associated with the absence of medium and high molecular weight vWF multimers, believed to be due to either impaired secretion of the large multimers or to excessive proteolysis of high molecular weight multimers. Mutations within amino acid residues 742-865 appear to cause structural changes in the molecule that interfere with intracellular polymerization or transport, or which make the multimers unusually sensitive to degradation by proteases.

Type 2B: variants with increased affinity for GPIb, a gain-of-function mutation. Thirteen missense mutations have been identified in this subtype, which are clustered within a short segment of the A1 domain. These point mutations in the A1 domain appear to cause the sort of conformational change that would exposed the normally cryptic binding site for GpIb.



vWF mutations associated with type 2B vWD phenotype clustered within the A1 domain of vWF (20)

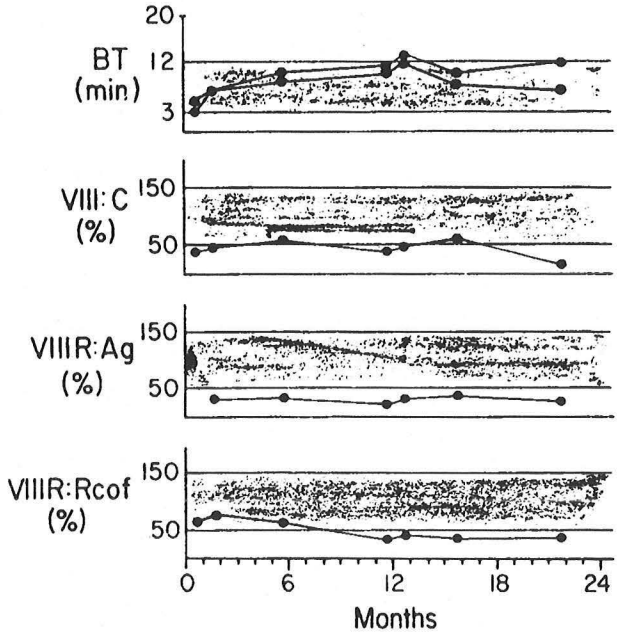
Type 2 M vWD refers to variants with decreased platelet-dependent function, not caused by the absence of high molecular weight multimers.

Type 2 N (N for Normandy, where the first case of this type was described) refers to qualitative variants where there is decreased affinity for factor VIII, usually associated with mutations in the factor VIII binding site. It may be difficult to separate from classical hemophilia, both clinically and in terms of laboratory studies[26].

Type 3 is quite uncommon and is manifested by virtual **absence of vWF;** Some cases are likely the result of homozygous (or doubly heterozygous) inheritance of abnormal vWF genes and was likely the abnormality seen in von Willebrand's index case. Fortunately, this severe type constitutes a very small number of patients, with an incidence of approximately 1:1,000,000[27, 28]. The incidence of this variant is actually much less than would be expected from autosomal recessive inheritance, given the known incidence of the Type I and II variants; some cases may be a result of mutations which affect mRNA transcription, processing or stability.

LABORATORY DIAGNOSIS OF VON WILLEBRAND'S DISEASE

It can be difficult to confirm the diagnosis of vWD, especially in mild cases. Usually a series of tests is required to make the diagnosis and classify the disease appropriately. Often it is necessary to perform a series of tests to make a diagnosis, as not all will be abnormal in the same subject at anyone time [29-31]. A number of physiologic and other stimuli increase von Willebrand factor levels, including high estrogen levels associated with oral contraceptive and pregnancy, inflammatory states, exercise and adrenergic stimuli and advancing age. It may often require repeated testing, even in the face of a suggestive bleeding history, to confirm the diagnosis of von Willebrand's disease [32].



Results of serial studies in a patient with Type 1 vWD (32)

A. General tests useful in diagnosing von Willebrand's disease

a. Bleeding time

This physiological test is part of the initial battery of tests in any patient whose symptoms suggest a defect in primary hemostasis. It is usually significantly prolonged in von Willebrand's disease. It can be useful to assess treatment effects. Standardization of the bleeding time is very difficult, however, and a mildly prolonged bleeding time do not necessarily connote an increased risk of surgical or other clinical bleeding, nor does a normal bleeding time rule out vWD [33, 34].

b. Activated partial thromboplastin time

If the factor VIIIC level is reduced to 25-30% or less of normal there is usually a modest prolongation of the activated partial thromboplastin time (aPTT), the usual screening test for the intrinsic coagulation pathway.

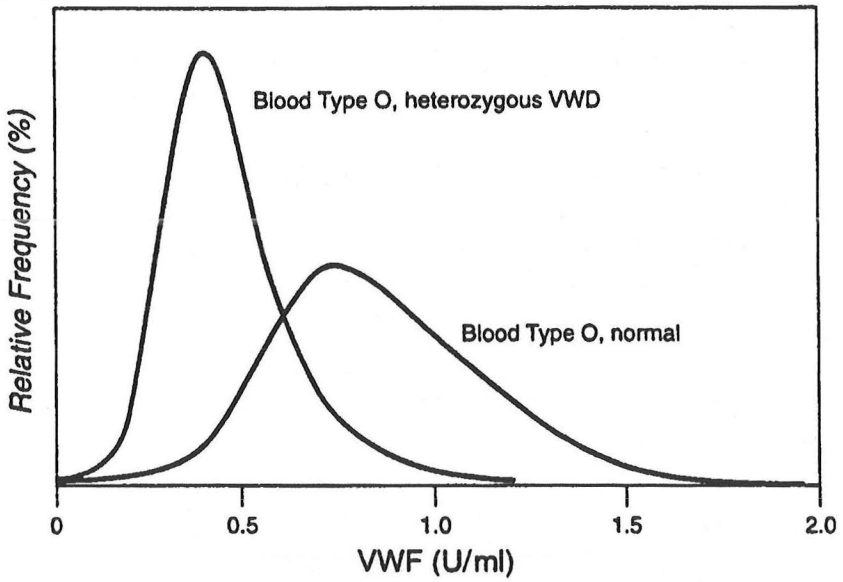
B. Tests which are more specific for von Willebrand's disease: see Table 1 for standard terminology [35]:

Table 1: Standard Terminology in von Willebrand's Disease
(from International Committee of Hemostasis and Thrombosis)

	Abbreviation	Definition	Measurement Technique
General Term			
Factor VIII (antihemophiliac factor)	FVIII (AHF)	Plasma protein deficient in patients with severe classic hemophilia (hemophilia A). Reduced, but not absent, in mild to moderate hemophilia.	
Immunologic Identity			
Factor VIII antigen	FVIII:Ag	Antigenic determinant (or determinants) on FVIII	Immunoassays employing human or monoclonal antibodies
Functional Identity			
Factor VIIIC	FVIII:C	Coagulant property of FVIII (often used interchangeably with FVIII)	Standard coagulation assays -aPTT
General Term			
von Willebrand factor	vWF	Large, multimeric glycoprotein necessary in vitro for normal platelet adhesion and in vivo for normal bleeding time	
Immunologic Identity			
von Willebrand factor antigen	vWF:Ag	Antigenic determinant (or determinants) on vWF. Previously called FVIII-related antigen (FVIIIR:Ag),	Immunoassays employing Heterologous antibodies to the FVIII/vWF complex
Functional Identity			
Ristocetin cofactor activity	RCoF activity	Ability of normal plasma to induce agglutination of washed or fixed normal platelets on exposure to the antibiotic ristocetin	Ristocetin cofactor assay (quantitative measure of vWF activity)

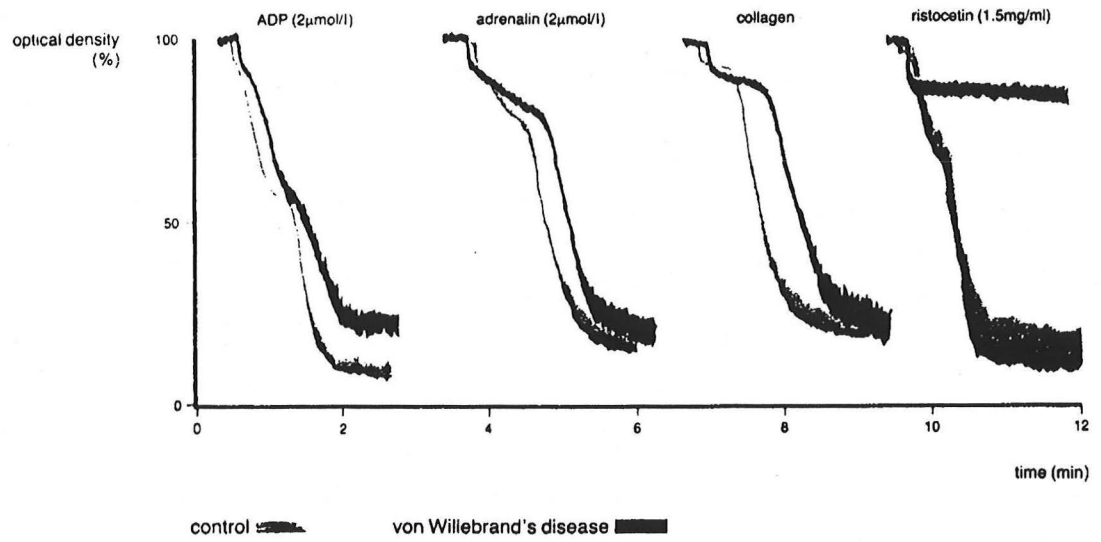
b. Von Willebrand factor antigen (vWFAg) - this can be measured in two ways, using either an ELISA or a Laurell rocket type analysis. Its value does not imply any functional activity. In Type 1 it will usually correlate closely with the functional activity, but in the type 2, with quantitatively abnormal vWD it is often significantly higher than the ristocetin cofactor activity.

c. Ristocetin cofactor assay (RCoF) - this is the most important functional assay of the vWF; it measures the ability of the vWF in the patient's plasma to aggregate normal formalinized platelets on addition of Ristocetin. Platelet aggregation can be induced in normal subjects by Ristocetin, an undesirable side effect of this otherwise valuable antibiotic, which caused its withdrawal from therapeutic use. However, this property has been a boon to understanding platelet function, especially in von Willebrand's disease. Ristocetin appears to expose the binding site on vWF for the platelet GpIb; binding this receptor initiates platelet activation and leads to platelet aggregation. Ristocetin cofactor activity parallels the severity of the disorder, but also is subject to fluctuations with inflammation, hormone therapy and other stimuli. Blood group alters its expression, both in normals and in vWD. Individuals of Blood group O have consistently lower levels, so that even normal individuals have RCoF levels that dip below the normal range [36].



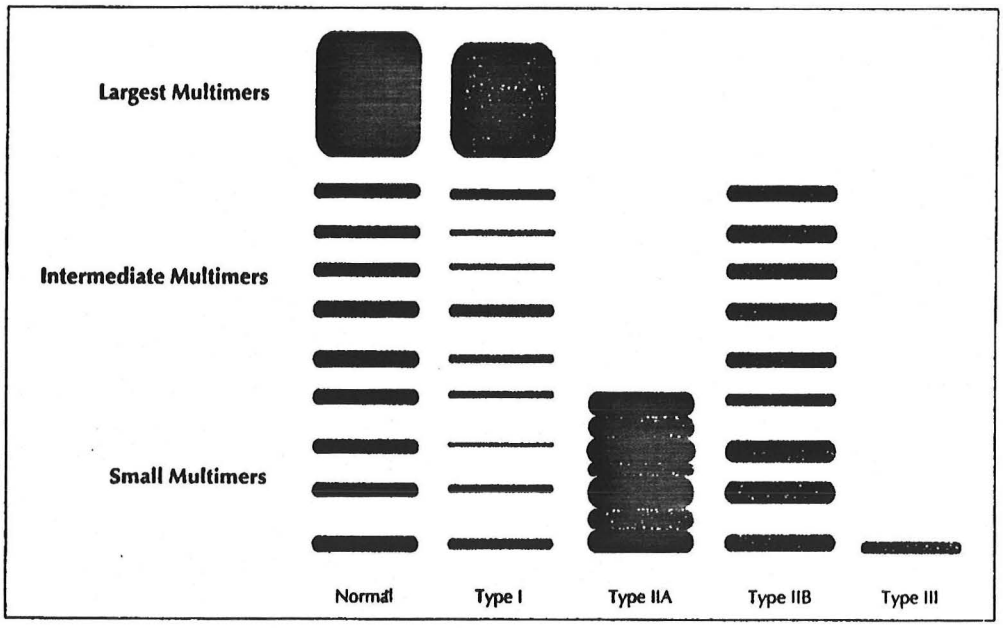
Distribution of VWF:Ag levels in normal donors of blood group O, compared with the range for patients heterzygous for vWF abnormality [36]

d. Ristocetin-induced platelet aggregation (RIPA) - this assay involves adding Ristocetin to a suspension of the patient's platelet-rich-plasma, and monitoring aggregation. In most variants of von Willebrand's disease platelet aggregation in response to Ristocetin is reduced. There is one important exception to this pattern: in the type 2B von Willebrand's disease where the Ristocetin induces increased aggregation of platelets. This is important clinically, especially in relation to using DDAVP in treatment of Type 2B.



Platelet aggregation studies to ADP, epinephrine, collagen and ristocetin in vWD, showing reduced aggregation to ristocetin

e. Multimer analysis - multimers analysis involves electrophoresis of plasma (or platelet lysate) on low concentration agarose or acrylamide/agarose gels. The multimer pattern is then visualized by labeling with an antibody to vWF. The highest weight multimers, which may be as large as 20,000 kilodaltons appear as a blur at the top of the gel, with increasingly clear bands below, corresponding to the smaller multimers. The pattern of multimers is very helpful in sub-typing the VWD [23]. There is a normal distribution of multimers in Type 1 vWD, although there is quantitative reduction; Type 2A shows loss of high and intermediate multimers, with extra bands in the low molecular weight range corresponding to excess proteolysis; type 2B shows loss of high molecular weight bands, which have boiund on to platelet receptors; type 3 shows virtual absense of multimers.



Multimer patterns in vonWillebrand's disease [16]

Table 2: Patterns of test results in von Willebrand's disease

Type	VIIIC	vWF antigen	RCoF activity	RIPA	Multimers
1	↓	↓	↓	↓	Normal pattern Reduced quantity
2A	N or ↓	N or ↓	↓↓	↓↓	Reduced high and intermediate multimers
2B	N or ↓	N or ↓	↓ or ↓↓	↑↑	Reduced high molecular weight monomers
2M	N or ↓	N or ↓	↓ or ↓↓	↓ or ↓↓	Normal
2N	↓↓	N	N	N	Normal
3	↓↓	↓↓	↓↓	↓↓	Absent

CLINICAL FEATURES OF VON WILLEBRAND'S DISEASE

Unlike von Willebrand's index case, the majority of patients with vWD have a relatively mild course. The defect in primary hemostasis produces a well known constellation of bleeding manifestations including: ready and spontaneous bruising; prolonged bleeding from cuts; mucous membrane bleeding, particularly epistaxis; menorrhagia and surgical bleeding. Gastro-intestinal bleeding is uncommon. Joint bleeding and deep tissue hemorrhage, characteristic of classical hemophilia, is extremely uncommon, except in the type II Normandy variant. As with many patients with defects in primary hemostasis, these may be dramatically worsened by ingestion of aspirin or non steroidal anti-inflammatory drugs (NSAIDS). Because of the early interest and high frequency of disorder in Scandinavian countries some excellent population studies have come from this area including a remarkable comprehensive monograph by from Silwer which detailed every known case in Sweden at the time, from which the following table is taken, complete with terminology [37].

Table 3

Frequency (%) of remarkable bleeding in the Swedish series (264 cases) of von Willebrand's disease (37)

Nose bleeding	62.5
Meno-metrorrhagia	60.1*a*
Post-extraction haemorrhage	51.5
Ecchymoses and haematomas	49.2
Bleeding from trivial sores and wounds	36.0
Gingival bleeding	34.8
Postoperative bleeding	28.0
Bleeding at delivery	23.3*a*
Gastrointestinal bleeding	14.0
Traumatic oral and lip bleeding	11.7
Petechiae	11.5
Joint bleeding	8.3
Haematuria	6.8
Ovarian bleeding	6.8*a*
Bleeding from tonsils	6.1
Bleeding during shedding of teeth	4.9
Bleeding at abortion	3.8
Intramuscular, deep subcutaneous or submucous bleeding	2.7
Bleeding from ears	3.0
Haemoptysis	1.9

a Calculated for females above 15 years.

A recent study of documented menorrhagia in Swedish women showed that 20 percent had laboratory studies consistent with mild von Willebrand's disease whereas the incidence of vWD in this general population is 1-2% [38].

The bleeding manifestations of von Willebrand's disease can vary significantly, both in the individual and in affected family members. In type 1 von Willebrand's disease there is mild to moderately severe bleeding problems. The inadequate levels of vWF in plasma is always associated with an equivalent decrease in factor VIII C activity and the symptoms essentially parallel the level of vWF. Type 2 von Willebrand's disease, where there is a defective molecule, has a very heterogeneous clinical spectrum, with moderately severe to severe bleeding diathesis. Type 3 von Willebrand's disease is the most severe and there are significant clinical defects in both primary and secondary hemostasis and a commensurately severe clinical picture

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TREATMENT OF VON WILLEBRAND'S DISEASE

Most patients with von Willebrand's disease need little or just intermittent treatment. The majority of patients have relatively minor symptoms, and treatment is only required in the setting of surgery or trauma [30, 39-41]. Effective therapy is available for all forms of inherited von Willebrand's disease.

Treatment falls into three basic categories.

1. Use of DDAVP to release of stored von Willebrand's disease.

2. Replacement of von Willebrand's factor with plasma-derived products.

3. Prevention of fibrinolysis.

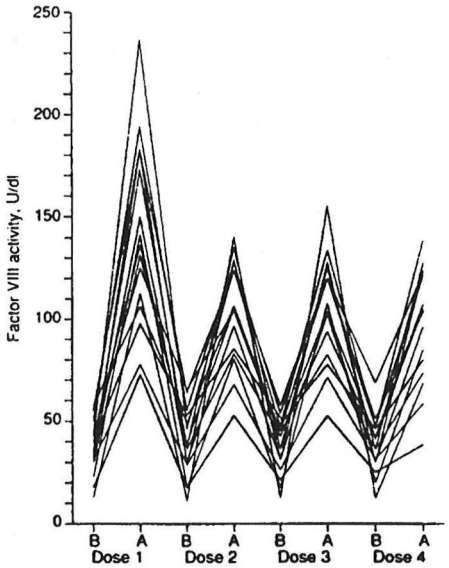
Other agents such as hormones for menstrual suppression may also be helpful in selected cases [42, 43].

1. DDAVP: - it was discovered serendipitously that high doses of the anti-diuretic vasopressin analog, 1-desamino-8-d-arginine vasopressin (DDAVP), caused release of factor VIII and von Willebrand's factor in normal individuals. It was immediately realized that this could have a very important role in treatment of mild hemophilia A and von Willebrand's disease, and studies quickly appeared documenting its role in this area [44-48].

There is a very rapid release of vWF from its endothelial cells storage sites (the Weibel-Palade bodies), and factor VIIIC also rises simultaneously. The bleeding time will often be dramatically reduced or even will normalize. DDAVP also stimulates release of tissue plasminogen activator (tPA). However, the hemostatic effect outweighs the fibrinolytic potential.

For patients with type I von Willebrand's disease a single dose of DDAVP will usually increase the von Willebrand factor two to three fold. Intravenous infusion

and subsequent doses were usually equivalent to the response to the second dose. Bleeding time similarly responded to daily doses; very little tachyphylaxis was seen with this physiological measurement.



Factor VIIIC activity in 15 patients with Type 1 vWD receiving daily DDAVP [49]

Intranasal DDAVP is a very important adjunct to treatment of von Willebrand's disease [50, 51]. It is a convenient route for self administration, and it comes in an easily administered multi-dose vial. Peak levels of vWF are not usually reached till 60 minutes after the inhalation, compared with 30 minutes for the intravenous form. It too shows approximately a threefold increase in factor VIII concentration and vWF, and the majority of patients show a shortening of the bleeding time.

Subcutaneous administration of DDAVP is also effective; like the nasal route it has a longer median time to peak increase, usually around 60 minutes. However, it eventually can give levels similar to those achieved the with intravenous route. Unfortunately, only a low concentration solution is available in this country, which makes this a less popular route.

The side effects of DDAVP are few. Most patients get facial flushing during the infusion, and occasionally patients complain of a mild headache. Because it is a powerful anti-diuretic agent there is a risk of hyponatremia, especially in patients with an electrolyte imbalance who are getting frequent infusions of DDAVP [52]. There have been concerns about the incidence of myocardial infarction in patients receiving DDAVP; most of the events noted occurred in elderly men who had other risk factors for arterial thrombosis, and there is no clear-cut evidence for the episodes reported being directly due to DDAVP. However, it is prudent to use it DDAVP cautiously in older individuals or individuals with coronary artery disease [53].

DDAVP appears most effective in type 1 von Willebrand's disease patients. This compound is ineffective in type 3 von Willebrand's disease and has variable effects in the type 2 forms [54]. There was a lot of concern about its use in type IIb von Willebrand's disease because of DDAVP-induced platelet aggregation and thrombocytopenia [55]. McKeown *et al* suggested that this may, at least in part, be an *in vitro* phenomenon; they did not observe reduction in patients' capillary platelet counts, although anticoagulated samples from the same individuals consistently demonstrated thrombocytopenia and platelet clumping [56].

2. Factor replacement with plasma derived products

Judith Pool's serendipitous discovery of cryoprecipitate, the precipitate rich in factor VIII and von Willebrand's factor which develops in slowly thawed plasma was a milestone in the treatment of hemophilia A and von Willebrand's disease. Until the last few years, cryoprecipate was the blood product of choice for patients with von Willebrand's disease [57-59]. Cryoprecipitate from ten blood donations was pooled for a single adult dose. Because the product cannot be standardized, factor levels needed to be monitored fairly closely. The major concern about this product is transmission of viruses, particularly hepatitis and AIDS.

The high purity factor VIII concentrates developed to eradicate AIDS and hepatitis viruses contain little useful vWF. However, some of the intermediate purity concentrates do contain significant amounts of von Willebrand factor, in the form of the high molecular weight multimers needed for primary hemostasis. Currently Humate-P appears to be the most effective of these products [60-63]. This product should be used when patients have inadequate response to DDAVP, when DDAVP is contra-indicated or when patients are having major surgery, such that they require higher levels than can be achieved by DDAVP.

These intermediate purity factor VIII products are not yet approved by the FDA for use in von Willebrand's disease; consequently vials do not show vWF content. However, it is generally accepted that they have an important role to play and they are part of American Society of Hematology's recommendations for von Willebrand's disease management - see Table 4. We usually dose patients on a twice daily basis, and monitor Ristocetin cofactor and factor VIIIC. It is possible to combine DDAVP and factor replacement to reduce the amount of these very expensive clotting concentrates.

In France a high purity von Willebrand's factor has been isolated from plasma (Facteur von Willebrand), but this is not yet available in the U.S [64] .

Table 4: Treatment strategies for von Willebrand disease, according to type
American Society of Hematology

VWD Variant	Hemorrhage	Therapy
Type 1	Minor	DDAVP[1]
	Major/surgery	Humate-P +/- DDAVP[2]
Type 3	Minor	Humate-P
	Major/surgery	Humate-P
Type 2A	Minor	DDAVP +/- Humate P[3]
	Major/surgery	Humate-P
Type 2M	Minor	DDAVP +/- Humate P[3]
	Major/surgery	Humate-P
Type 2B	Minor	DDAVP +/- Humate P[3]
	Major/surgery	Humate-P
Type 2N	Minor	DDAVP +/- Humate P[3]
	Major/surgery	Humate-P

1 The use of DDAVP in any of the above clinical settings in dependent on the response seen. An adequate response results in vWF levels of 40-50 U/dl or normalization of the bleeding time at 1 hr after infusion. All individuals should have a documented response before the therapeutic use of DDAVP is considered.

2 The use of vWF containing FVIII concentrates can be reduced in this setting by the use of DDAVP for 3-7 days following 3 days of therapy with concentrate.

3 The use of DDAVP in these clinical settings may result in a transient response that may be clinically efficacious depending on the nature of the bleeding episode. For individuals with no response to DDAVP or for more serious events in those with a documented response, vWF containing FVIII concentrates should be used.

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3. Fibrinolytic Inhibitors

Fibrinolytic inhibitors such as ε-amino caproic acid (Amicar) and tranexamic acid are often a useful treatment adjunct when there is mucosal bleeding or dental surgery in these patients. It was initially felt that, because DDAVP also increased tPA, that fibrinolytic inhibitors should be part of routine management in patients receiving DDAVP. However, this has not been found necessary and we reserve these agents for patients at risk of mucosal bleeding. It is important to be very cautious of their use in patients with liver disease, such as post transfusion hepatitis C. Here reduced clearance of activated clotting factors may predispose to low grade chronic DIC; inhibiting the protective fibrinolytic response may make this DIC clinically significant.

ACQUIRED VON WILLEBRAND'S DISEASE

This is usually an adult onset bleeding diathesis with a clinical history suggestive of a defect in primary hemostasis, such as bruising, mucous membrane bleeding, epistaxis or post operative bleeding: essentially the same pattern seen in patients with the inherited form of von Willebrand's disease [65, 66]. Laboratory studies in such patients show that the functional von Willebrand's factor measurement, the Ristocetin cofactor activity, is low and sometimes almost absent, even though the antigenic form of von Willebrand's factor may be normal or just mildly reduced. The bleeding time is prolonged. The multimeric pattern of von Willebrand's factor is similar to type 2A vWD, where there is a selective loss of the high and intermediate molecular weight multimers.

Acquired vWD occurs in a number of different clinical settings, see Table 4. By far the most common is the association with lymphoproliferative disorders or plasma cell dyscrasias, especially with parpaprotein (monoclonal protein) formation These represent more than 50 percent of the reported cases. Most of the time paraproteins are IgG, and about half of the implicated plasma cell dyscrasias fall into the category of monoclonal gammopathy of unknown significance (MGUS). Another association is with auto-immune disorders, particularly hypothyroidism, but also lupus and sclerodurma.

Acquired vWD is also seen in myeloproliferative disorders, particularly essential thrombocythemia and polycythemia vera. In this setting the level of vWF is usually inversely related to the cell counts, i.e., the higher the platelet count the lower the level of vWF. When platelet counts are controlled the VWF level will return to normal.

Other cases have been noted in the setting of tumors, particularly Wilm's tumor, adrenal cell carcinoma and other adenocarcinomas [67]. One study of 50 patients with Wilm's tumor showed that four (8%) had acquired von Willebrand's disease. In those cases associated with malignant disorders, the acquired vWD resolves if the tumor is successfully removed. Sporadic cases have been reported in association with drugs, including ciprofloxacin [68], griseofulvin, hydroxyethyl starch (HES) and valproic acid.

PATHOPHYSIOLOGY OF ACQUIRED VON WILLEBRAND'S DISEASE

It is believed that there are two fundamental mechanisms for acquired von Willebrand's disease:

1. Antibody binds to von Willebrand's factor, probably to a nonfunctional domain on the multimer. In the case of the plasma cell dyscrasias it appears that the paraprotein has this type of antibody activity. This immune complex is then cleared very rapidly by the reticulo-endothelial system. It is unclear why the high molecular weight multimers are preferentially cleared.

Table 5: Disorders Associated With Acquired von Willebrand's Disease and Their Approximate Frequencies [65]

Monoclonal gammopathy of unknown significance	27%
Multiple myeloma	15%
Non-Hodgkin's Lymphoma	10%
Myeloproliferative disorders	10%
Wilm's tumor	8%
Drugs	7%
Chronic lymphocytic leukemia	5%
Hypothyroidism	5%
Carcinoma	3%
Hairy-cell leukemia	2%
Unknown	8%

Unlike the traditional coagulation inhibitors, the Ristocetin co-factor inhibitors of acquired von Willebrand's disease are corrected by addition of normal plasma. It is believed that this is evidence that the antibody is binding to a nonfunctional area on the von Willebrand's protein, and it is predominantly the enhanced clearance that reduces the vWF.

2. Absorption of von Willebrand factor to tumors or abnormal cells such as the abnormal platelets in the myeloproliferative disorders. Some studies have shown immunofluorescent staining of von Willebrand's factor within tumor cells. There are several case reports of the acquired von Willebrand's disease resolving after successful removal of a tumor or treatment of the underlying myeloproliferative disorder..

TREATMENT

Treatment of acquired von Willebrand's disease depends on the underlying cause. Replacement of vWF with cryoprecipitate or Humate-P is effective in the short-term, but the active component of the factor is rapidly cleared. Similarly, when DDAVP is used to cause vWF secretion, this will be cleared more rapidly than usual. There are reports of successful treatment with intravenous immunoglobulin infusion in the cases associated with paraproteins in which the vWF is part of immune complex [69]. This treatment is often dramatically effective in the short-term, the response varying from one to three weeks. Some reports show the use of monthly IVIG maintenance to maintain a normal ristocetin co-factor level can prevent the bleeding manifestations.

When the disorder is presumed to be caused by a lympho-proliferative or plasma cell proliferative disorder, appropriate treatment of the underlying malignancy may suppress the paraprotein abnormality. In the acquired von Willebrand's disease associated with monoclonal gamopathy of undetermined significance, pulse dexamethasone has been found to be effective. Myelosuppressive therapy to control thrombocythemia will help control the syndrome secondary to myeloproliferative disease. Surgical removal of implicated tumors has similarly been found to reverse the syndrome.

SUMMARY

Von Willebrand's disease was first described seventy years ago. Since then there has been remarkable progress in understanding the disorder and devising effective treatment. Serendipity has been more than usually important in this progress, from the discovery of the mechanism of Ristocetin-induced aggregation of platelets, or lack thereof in vWD, to Judith Pool's accidental discovery of cryoprecipitate and the later unexpected effect of DDAVP on endothelial cell secretion of vWD. These leaps, plus more traditional progress has given us knowledge of the molecular structure which forms the basis for a rational classification, in which laboratory studies can usually readily diagnose and subtype the disease, for which effective and safe treatment is available.

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