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Effectiveness of High-Dose Intravenous Immunoglobulin in a Case of Acquired von Willebrand Syndrome With Chronic Melena Not Responsive to Desmopressin and Factor VIII Concentrate

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A patient with benign monoclonal IgG lambda paraproteinemia, acquired von Willebrand syndrome (AVWS), and chronic melena successfully responding to high-dose intravenous immunoglobulin (IvIg) is reported. Coagulation parameters at admission were APTT (ratio) 1.68; VIII:C 11 IU/dL; vWF:Ag 7 IU/dL: Ricof <3 IU/dl. RIPA was >1.8 mg/ml, and bleeding time (BT) was prolonged (18 min). No evidence for an in vitro inhibitor against the VIII/vWF complex was observed. VIII/vWF measurements showed a short-lived increase after both DDAVP and Hemate P, and BT was transiently normalized. After intravenous Ig (1 g/kg for 2 days), VIII/vWF measurements, hemostatic parameters and multimeric pattern were completely corrected (VIII/C 106 IU/dl, vWF:Ag 168 IU/dl, RiCof 147 IU/dl, APTT ratio 0.89, BT 5′), with a return to pre-infusion values after 15 days. Hemoccult test became negative and packed red cell transfusions, of which 130 units were administered during the last year, were no longer required. After 18 months the patient is on maintenance treatment with repeated courses of Ig, at 3 to 4-week intervals based on VIII/vWF and BT monitoring.

Key words: acquired von Willebrand syndrome, intravenous immunoglobulin, von Willebrand factor

INTRODUCTION

The acquired von Willebrand syndrome (AVWS) is a rare coagulopathy, closely resembling inherited von Willebrand disease, mostly associated with lympho or myeloproliferative disorders or monoclonal gammopathies [1,2]. Patients with AVWS may present with severe hemorrhagic episodes of difficult management. Recently, high-dose intravenous immunoglobulins (IvIg) have been reported to improve factor VIII/von Willebrand factor (VIII/vWF) levels in two patients with AVWS, leading to successful prevention of bleeding during surgery and tooth extraction [3].

We describe a new case of AVWS with life-threatening recurring melena poorly responsive to DDAVP and factor VIII concentrate successfully treated during 18 months with repeated courses of high-dose IvIg.

CASE REPORT

A 66-year-old man with benign monoclonal IgG lambda paraproteinemia and AVWS was referred for chronic melena. On admission, physical examination was unremarkable apart from severe pallor (hemoglobin 7.7 g/dl). The patient had received 130 units of packed red cells during the previous 13 months during several admissions to a peripheral hospital. Several repeated instrumental examinations failed to demonstrate any ulcerative or neoplastic lesion, despite bloodlike material was repeatedly found in the caecum. Scintigraphy with ^{99m}Tc-labeled red cells showed two positive foci in the right lower abdomen, just below the inferior margin of the kidney, suggesting angiodysplasia. Hemoccult test was consistently, strongly positive (+++).

At admission, the complete blood count, bone marrow aspirate, and biochemical tests were all unremarkable. Immunoelectrophoresis revealed in IgG lambda mono-

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clonal protein (IgG 930 mg/dl). APTT (ratio) was 1.68; factor VIII procoagulant activity (VIII:C) 11 IU/dl; vWF antigen (vWF:Ag) 7 IU/dl; Ristocetin Cofactor (RiCof) <3 IU/dl. Ristocetin-induced platelet agglutination (RIPA) was severely impaired (>1.8 mg/ml) and bleeding time (BT) prolonged (18 min).

Desmopressin (DDAVP; Minirin, Valeas, Milan) and Hemate P (Behring, Marburg, Germany) were infused with limited clinical benefit (see Results). A trial for 6 months with azathioprine (150 mg/day) was started before Ig treatment but was without effect on laboratory parameters and transfusional requirement. Intravenous Ig (1 g/kg for 2 days) was then tried.

MATERIALS AND METHODS

Blood Sampling and Coagulation Tests

Blood was anticoagulated with 3.8% sodium citrate (1:10 v:v) and centrifuged at 1,500g for 15 min. In samples harvested to analyze the multimeric pattern of vWF, 5 mM EDTA, 6 mM N-ethylmaleimide, and 1 mM leupeptin (Sigma, St. Louis, MO) were added to the citrate anticoagulant to avoid in vitro proteolytic degradation of vWF after venipuncture.

BT was measured with the Simplate II device (General Diagnostics, Morris Plains, NJ). VIII:C, vWF:Ag, and RiCof were assayed as previously described [4,5]. RIPA was evaluated by adding increasing amounts of ristocetin to platelet-rich plasma and recording the dose required to obtain at least 30% of aggregation after 3 min.

The multimeric structure of plasma vWF was analyzed by Western blotting and visualization of the multimeric pattern using peroxidase-conjugated rabbit anti-vWF antibodies according to the method of Tomita et al. [6].

Platelet vWF was measured after platelet separation on Ficoll-Hypaque gradients [7]. Lysis was obtained by adding 1/40 vol of Triton X-100. After centrifugation at 3,500*g* at 4°C for 10 min, the supernatants were snap-frozen at −80°C for vWF:Ag and RiCof assays.

All VIII/vWF measurements were expressed as International units (IU), with reference to an internal plasma pool standardized against the 2nd International Reference Preparation for factor VIII/von Willebrand factor-related activities.

Inhibitor Studies

The IgG fraction of the patient and normal controls were purified by affinity chromatography on Sepharose–Protein A (Pharmacia, Uppsala, Sweden).

Mixing study. Normal plasma was mixed in a 1:1 ratio with the subject plasma and incubated for 2 hr at 37°C. The samples were then centrifuged at 2,500*g* for 15 min and VIII/vWF measurements assayed.

vWF-binding antibody assay. The technique of Fricke et al. [8] was used, with minor modifications. In this assay, serially diluted IgG purified from patient and normal plasma are bound to protein A and incubated with a constant amount of normal plasma. Residual VIII/vWF activities are then measured on the supernatant of the adsorbed plasma.

Inhibition of vWF binding to collagen. Binding of normal plasma vWF to collagen, in the presence of normal or patient purified IgG was studied using the enzyme-linked immunosorbent assay (ELISA) method of Brown and Bosak [9].

RESULTS

Laboratory Investigations

The analysis of the multimeric pattern of plasma vWF revealed a marked reduction of larger multimers (Fig. 1, lane 2). Platelet vWF:Ag (38 IU/10^9 plt; normal range 0.19–0.62 IU/10^9 plt) and RiCof (30 IU/10^9 plt; normal range 0.22–0.88 IU/10^9 plt) were normal. No evidence of inhibitory activity against any of VIII/vWF measurements was found by mixing studies or by vWF-binding Ab assay or inhibition of vWF binding to collagen (data not shown).

Table I shows the results of VIII/vWF measurements and BT before and after DDAVP infusion (0.4 μg/kg). BT was normalized at the end of infusion but again became prolonged at 2 hr. Only a very slight, short-lived increase of VIII/vWF measurements followed the DDAVP infusion. Six closely spaced infusions (at about 12 hr intervals) were administered, but tachyphylaxis rapidly ensued with no increase of VIII/vWF measurements after the third infusion. Hemoccult test remained strongly positive (+++).

Table II shows the results of VIII/vWF measurements and BT after Hemate P infusion (40 U/kg). Also in this case BT was shortened 30 min after infusion, but again prolonged at 6 hr. Larger multimers appeared after both treatments, but for a very short period of time (data not shown).

After IvIg, VIII/vWF measurements and hemostatic parameters were completely corrected (VIII/C 106 IU/dl, vWF:Ag 168 IU/dl, RiCof 147 IU/dl, APTT ratio 0.89, RIPA 0.9 mg/ml, BT 5 min). After 15 days a return to pre-infusion values was observed (Fig. 2). Multimeric analysis of plasma vWF showed the appearance of all multimers 24 hr after Ig infusion and their progressive clearance in the following 15–21 days (Fig. 1, lanes 3–6). Hemoccult test became negative, and no packed red cells were further required. Repeated courses with a single infusion (1 g/kg for 1 day) proved similarly effective in improving VIII/vWF measurements and BT with return to basal level in about 3–4 weeks (Fig. 3). On the basis of laboratory monitoring of VIII/vWF measurements, a total of 24 Ig infusions were administered every 3–4 weeks and blood transfusions were no longer needed during the

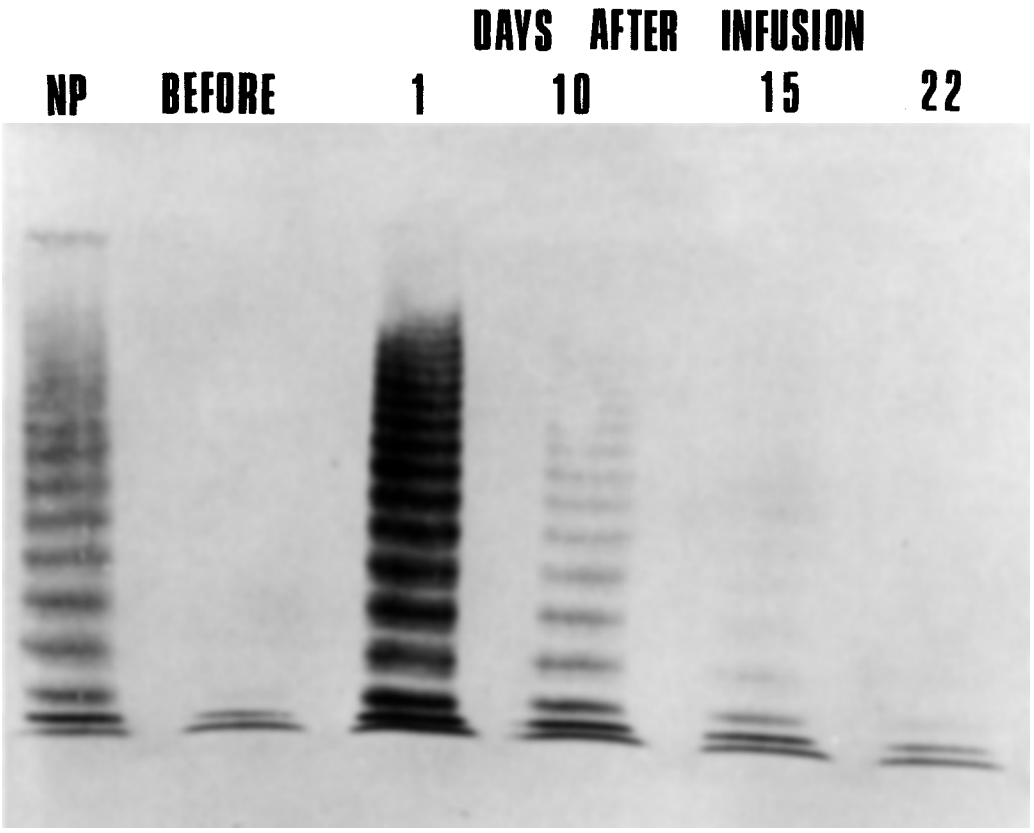

Fig. 1. Multimeric pattern of plasma von Willebrand factor before and after the first immunoglobulin infusion. From left: lane 1, normal plasma; lane 2 patient plasma at baseline; lane 3–6 patient plasma after 1, 10, 15, and 22 days.

TABLE I. Bleeding Time and VIII/vWF Measurements Before and After First DDAVP Infusion

Time	VIII:C (IU/dl)	vWF:Ag (IU/dl)	RiCof (IU/dl)	Bleeding Time (min)
Before	13	8	<3	18
30 min	60	31	41	5.5
60 min	40	22	17	—
2 hr	18	12	<3	17
4 hr	16	10	<3	—

TABLE II. Bleeding Time and VIII/vWF Measurements Before and After Hemate P

Time	VIII:C (IU/dl)	vWF:Ag (IU/dl)	RiCof (IU/dl)	Bleeding time (min)
Before	12	8	<3	17
10 min	73	147	131	5.5
30 min	61	96	68	—
3 hr	17	20	7	—
6 hr	17	17	<3	>20
24 hr	12	9	<3	—

following 18 months. The patient is still on Ig treatment with a stable hemoglobin level around 15–16 g/dl.

DISCUSSION

Acquired von Willebrand syndrome has often been observed in patients with monoclonal gammopathy [1,3,10–12]. In rare instances, an antibody with inhibitory activity against one or more of the VIII/vWF measurements has been demonstrated [8,12,13]. The presence of autoantibodies binding to VIII/vWF complex and leading to the formation of a macromolecular complex rapidly cleared from circulation has also been rarely reported [14,15]. We were not able to demonstrate any inhibitory activity "in vitro" of the monoclonal protein of our patient. However, the rapid clearance of VIII/vWF after DDAVP and Hemate P infusion strongly supports the possibility of an antibody that "in vivo" binds to VIII/vWF. We have also ruled out the presence of an antibody inhibiting the binding of vWF to collagen, recently described in a patient with AVWS [16].

Patients with AVWS may often have only mild bleeding diathesis. In some cases, DDAVP proved successful in controlling or preventing hemorrhages [17,18], but usually VIII/vWF elicited post-infusion showed a more rapid decay than in congenital type I von Willebrand disease [1]. In our patient, only a very short-lived in-

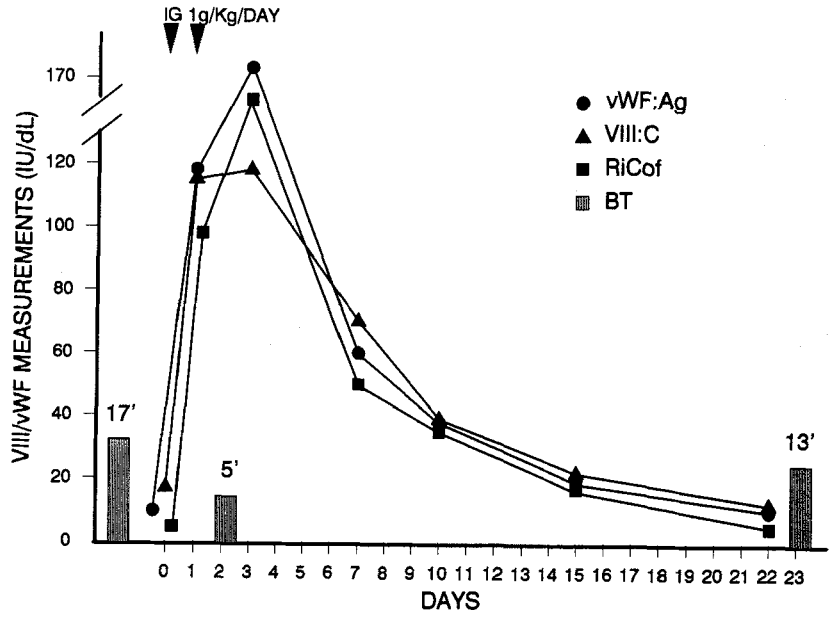

Fig. 2. VIII/vWF and bleeding time after the first immunoglobulin infusion.

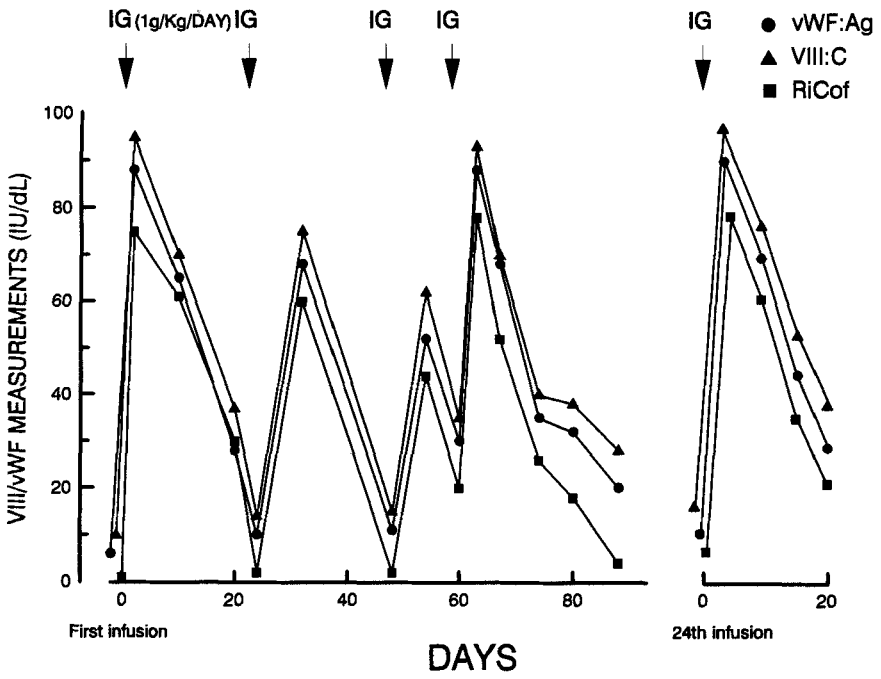

Fig. 3. Consistency of response to IvIg after the first course of immunoglobulin and after 24 courses.

crease was observed, with no clinical benefit. Other treatments, including large doses of cryoprecipitate [3,19,20] and plasmapheresis [21], have been also attempted. A factor VIII concentrate (Hemate P), particularly rich in vWF, induced a short-lived increase of VIII/vWF measurements in our patient, but very large amounts were required to obtain a reduction of packed red cell requirement. Recently, Macik et al. [3] reported the successful use of high-dose IvIg in two patients, one with monoclonal gammopathy and one with splenic B-cell lymphoma. In the first patient, the effect on VIII/vWF measurements lasted for about 21 days, allowing multiple dental extractions to be safely carried out. The second patient underwent diagnostic splenectomy under Ig. VIII/vWF measurements remained normal for 6 months after the single course of Ig, suggesting that this sustained

effect could probably be due to the removal of the neoplastic spleen. In our patient, Ig infusion induced a prompt and sustained normalization of VIII/vWF measurements and BT for 15–21 days. On the basis of follow-up assay of VIII/vWF measurements, repeated courses of Ig were administered (Fig. 3), without recurrence of bleeding. To our knowledge, another three similar cases, reported only in abstract or letter form, have been described [22–24]. One of these patients became refractory after the successful rise of RiCof obtained with the first course of IvIg [24].

The mechanism of action of IvIg has not yet been determined. Some authors suggest the presence of anti-idiotype antibodies in the Ig preparations [25–27]. Alternatively, the blockade of Fc-receptors on the reticulo-endothelial system [28,29] or the elimination of circulating immune complexes by monomeric immunoglobulins [25] have been suggested.

In conclusion, this report confirms that the infusion of high-dose IvIg may be effective in improving VIII/vWF measurements and BT in AVWS, even in cases not responsive to DDAVP or FVIII concentrate. In this patient, Ig infusion led to the control of chronic melena with sustained increase of VIII/vWF measurements after each of the 24 courses administered during a period of 18 months.

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