BRIEF REPORT

Correction of the Hemostatic Defects in Von Willebrand's Disease

By Herbert A. Perkins

VON WILLEBRAND'S DISEASE (VWD) is a congenital bleeding tendency characterized by a long bleeding time, defective platelet adhesiveness, deficient plasma Factor VIII (antihemophilic factor), and a progressive rise in Factor VIII levels following appropriate transfusion therapy. The long bleeding time appears to be corrected by a specific plasma factor; it seems reasonable that this plasma factor is required for normal platelet adhesiveness and, thus, for formation of the platelet plug. The progressive rise in Factor VIII following transfusion of normal plasma into patients with VWD has been interpreted as evidence that these patients lack a factor, which here will be referred to as the "AHF stimulator," required for synthesis of the antihemophilic factor (AHF). The plasma factor which corrects the long bleeding time will be referred to as the "bleeding time factor." Although studies indicate that von Willebrand's disease is due to a single genetic defect, the AHF stimulator and the bleeding time factor appear to be physically distinct. The bleeding time factor has been demonstrated in fresh or fresh-frozen plasma and in the Blombäck I-O fraction, but not in fibrinogen concentrates, stored plasma, or Cohn Fraction I concentrates prepared in this country. In contrast, the AHF stimulator is present in fibrinogen concentrates and in serum.

Appropriate transfusion therapy can accomplish three beneficial effects for patients with von Willebrand's disease. The bleeding time is corrected, Factor VIII is transfused, and the patient is stimulated to increase his own level of AHF still further. Because of the last effect, it is obviously much easier to raise and maintain the Factor VIII level in the plasma of a patient with von Willebrand's disease than in a patient with classical hemophilia. Correction of the bleeding time has been more difficult to achieve. Nilsson and co-workers have shown that correction of the Duke bleeding time in an adult with Blombäck Fraction I-O generally requires a dose prepared from 1500 ml. of

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375

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plasma. The more sensitive Ivy bleeding time is usually not corrected by this therapy. Fortunately, the degree of correction manifested by a normal Duke bleeding time appears to provide adequate hemostasis for surgery; this undoubtedly explains Biggs' report that surgery could be safely performed if the AHF was elevated even though the (Ivy) bleeding time was not corrected. In a later communication, she pointed out that her statement was not intended to apply to the situation where spontaneous bleeding had to be controlled. Weiss demonstrated that the Ivy bleeding time could be shortened by large volumes of very fresh plasma prepared to avoid glass contact, or by the Blombäck I-O Fraction, but not by plasma stored for a short period, by glass-contacted plasma, nor by Cohn Fraction I prepared by the Boston Protein Foundation or by Merck-Sharp and Dohme. It thus appears that the bleeding time factor is extremely labile and possibly lost on contact with glass. An obvious need exists for a readily available stable preparation of the bleeding time factor which is effective in a small volume. The present report was stimulated by the discovery that a Factor VIII concentrate prepared in the blood bank by readily available technics (Pool cryoprecipitate) is a reliable preparation for correction of the bleeding time in von Willebrand's disease, whereas newly available potent commercial AHF concentrates are not.

**MATERIALS AND METHODS**

1. Cryoprecipitate was prepared by the method of Pool and Shannon. One unit refers to the amount prepared from the plasma of a 500 ml. donation of blood.

2. Antihemophilic factor concentrates more potent than any previously available in this country were obtained from two sources: (1) Hyland Laboratories, Los Angeles, California; (2) Dr. Alan Johnson, American National Red Cross Research Laboratory, New York City.

3. Bleeding times were measured by the technic of Ivy in a deliberate attempt to use a test as sensitive and difficult to correct as possible.

4. Platelet adhesiveness was measured by the technic of Salzman. It might be anticipated that this should be reversed to normal along with the bleeding time, but Strauss and Bloom have reported that it does not change. Our experience with this test has convinced us that it is sufficiently reproducible to be useful in the diagnosis of von Willebrand's disease but that quantitative reproducibility is so poor that it is useless as a method for evaluating the effects of transfusion therapy.

5. Antihemophilic factor levels were assayed by a thromboplastin generation technic modified from Pool and Bergna. In our experience the Factor VIII levels achieved in patients given plasma or AHF concentrates of any type agree with those expected from the assay levels and volumes of the infused materials and patients' plasma.

6. Most of the transfusion experiments were carried out at times when the patients were not actively bleeding. The tables and figures contain data obtained only with patient F.P., since experience with patient W.W. was limited to a single transfusion episode.

**CASE REPORTS**

**Case 1**

F.P., a 30-year-old white male, was first admitted to the University of California Medical Center in San Francisco on May 28, 1964, with a 15 month history of massive gastrointestinal bleeding. This was the first such episode, although his previous history included bleeding of the gums as an infant when he nursed, one hospitalization for severe epistaxis, and prolonged bleeding after tooth extractions and trauma to the skin.

Tests in our laboratory at times when no transfusions had been given for a week revealed
CORRECTION OF HEMOSTATIC DEFECTS IN VWD

a bleeding time over 30 minutes, 0 per cent platelet adhesiveness, and AHF levels which varied between 25 and 45 per cent.

During the next 1\(\frac{1}{2}\) years, repeated gastrointestinal bleeding required 14 admissions (approximately 50 per cent of his days were spent in the hospital) and administration of nearly 1000 units of whole blood or certain of its components. Three attempts at surgical correction, culminating in removal of 10 feet of small bowel, failed to decrease the frequency or severity of gastrointestinal bleeding. The resected bowel contained multiple blue swellings from 1 to 3 mm. in size extending from the serosa to the mucosa. On microscopic examination, these were interpreted to be either hematomata or areas of phlebectasia. Bleeding was fairly well controlled during surgery by fresh whole blood and fresh-frozen plasma, but episodes of "spontaneous" bleeding occurred more frequently than usual in the following weeks.

Following transfusion therapy, his plasma AHF was always at a level associated with normal hemostasis, but his bleeding time was rarely and inconstantly corrected only when his rate of bleeding permitted administration of 6 to 8 units of fresh whole blood and fresh-frozen plasma within a relatively few hours. Various concentrates of antihemophilic factor were then administered because of the evidence that the bleeding time factor appears to be concentrated in at least one Factor VIII preparation (Blomback I-O). The results of these infusions will be detailed later in this paper.

Case 2

W.W., a 52-year-old man, was admitted on May 4, 1966, to the University of California Medical Center for evaluation of the effects of transfusion therapy. He had bruised easily all of his life and bled excessively after tooth extractions. Both he and his sister had had repeated episodes of gastrointestinal bleeding despite normal x-rays. His bleeding time was over 30 minutes, platelet adhesiveness was 7 per cent, and his Factor VIII level was 43 per cent.

EXPERIMENTAL RESULTS

In four early studies it was noted that correction of the bleeding time was more complete 4 hours after the experimental transfusion than it had been at 10 minutes following the infusion (Table 1). The effect of further trials on the bleeding time was therefore estimated by determining the bleeding time 4 hours after the transfusions.

The data obtained with two commercially prepared AHF concentrates are shown in Table 2. Both were obviously potent sources of AHF, but the effect on the Ivy bleeding time was negligible. (The apparent beneficial effect of three units of ARC material was not borne out by tests with 5 and with 10 units, and may be the result of the vagaries of this very imperfect test or a summating effect on top of fresh-frozen plasma given several days before the first infusion.)

In contrast, a dose of 8 units of cryoprecipitate (80 ml.) repeatedly and significantly shortened the bleeding time (Fig. 1), although not always completely into the normal range. Similar results were obtained with patient W.W. Evidence for lability of the bleeding time factor is presented on the right side of the figure. Eight units of precipitate from plasma stored at \(-20\) C. for 6 months were ineffective. Precipitate prepared from plasma obtained from blood stored for 21 days at 4 C. had less effect than did any of the infusions with cryoprecipitate prepared from fresh plasma, even though twice the usual dose of "aged plasma cryoprecipitate" was used.

A limited amount of evidence was obtained showing that some corrective
Table 1.—Effect of Time Interval Following Therapy on Correction of Bleeding Time

<table>
<thead>
<tr>
<th>Experiment No.</th>
<th>Therapy</th>
<th>Time Interval before Therapy</th>
<th>Bleeding Times</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>24 hr.</td>
<td>10 min.</td>
</tr>
<tr>
<td>1</td>
<td>3 U. ARC Concentrate</td>
<td>14'</td>
<td>11'</td>
</tr>
<tr>
<td>2</td>
<td>8 U. Cryoprecipitate</td>
<td>&gt;30'</td>
<td>11'</td>
</tr>
<tr>
<td>3</td>
<td>8 U. Cryoprecipitate</td>
<td>6'</td>
<td>14.5'</td>
</tr>
<tr>
<td>4</td>
<td>8 U. Cryoprecipitate</td>
<td>23'</td>
<td>&gt;30'</td>
</tr>
</tbody>
</table>

Table 2.—Effects of Commercial AHF Concentrates

<table>
<thead>
<tr>
<th>Date</th>
<th>Source</th>
<th>No. of Units</th>
<th>AHF Assays on Patient Plasma</th>
<th>Ivy B.T.</th>
</tr>
</thead>
<tbody>
<tr>
<td>6/12/64</td>
<td>ARC†</td>
<td>3</td>
<td>41% before infusion</td>
<td>14'</td>
</tr>
<tr>
<td>6/22/64</td>
<td>ARC</td>
<td>3</td>
<td>41% before infusion</td>
<td>14'</td>
</tr>
<tr>
<td>6/25/64</td>
<td>ARC</td>
<td>10</td>
<td>48 before infusion</td>
<td>&gt;30</td>
</tr>
<tr>
<td>6/15/65</td>
<td>Hyland‡</td>
<td>4</td>
<td>29 before infusion</td>
<td>29</td>
</tr>
<tr>
<td>9/20/65</td>
<td>Hyland</td>
<td>6</td>
<td>62 before infusion</td>
<td>100</td>
</tr>
</tbody>
</table>

* There was no recent therapy, but massive gastrointestinal blood loss had occurred.
† ARC unit consisted of a vial of lyophilized material which was dissolved in 100 ml. diluent. Average AHF assay value at time of transfusion was 180 per cent.
‡ Hyland unit (also lyophilized) was dissolved in 30 ml. diluent. Average AHF assay was 400 per cent.

Since the demonstration that cryoprecipitate will shorten his bleeding time, patient F.P. has been given 8 units of cryoprecipitate whenever melena recurs (at intervals of 4 to 8 weeks). When this has been done within 24-48 hours after black stools were noted, bleeding has ceased and hospital admission has been avoided. On several occasions, when for personal reasons he delayed hospitalization for 3 to 5 days before reporting for transfusion, repeated infusion of cryoprecipitates and red cell transfusions were necessary. The ability of early treatment with cryoprecipitate to eliminate need for red cell transfusion is particularly important in this case, since he has developed isoantibodies to two red cell factors (rh* and Kell) and to white blood cells.

DISCUSSION

These studies demonstrate that the Pool cryoprecipitate, a material now available in most large blood banks, is an effective source of the bleeding time factor lacking in patients with von Willebrand's disease. Corroboration of this fact has recently been obtained from England. Control of severe instances of spontaneous bleeding in Case 1 clearly required correction of the bleeding time; adequate levels of Factor VIII were not enough. It should be pointed
out, however, that most patients with von Willebrand's disease do not bleed severely enough to require transfusion therapy of this sort.

The lack of the bleeding time factor in new commercially prepared AHF concentrates, despite their very high levels of AHF, is disappointing, since it appears that even if these concentrates become available in sufficient quantity and at a low enough price to be the preferred therapy for hemophilia, cryoprecipitates or other preparations containing the bleeding time factor will still be needed for patients with von Willebrand's disease.

The obvious lability of the bleeding time factor creates handicaps in the preparation of concentrates. The success of the Pool cryoprecipitate may lie in the protection provided by rapid processing of a single unit at a time—all at cold temperatures. The all-plastic system may be important, but final proof is still lacking. We transferred one batch of dissolved cryoprecipitate to an uncoated glass bottle, rotated it over the glass surface for one minute, and then transfused it. The bleeding time correction was in the usual range. Weiss's report, however, indicates that glass contact may explain in part the poor results with most AHF concentrates.
Summary

The Pool cryoprecipitate provides an effective source of the factor lacking in the plasma of patients with von Willebrand’s disease which is necessary for a normal bleeding time. Correction of the bleeding time appears to be necessary for control of bleeding in some instances of severe spontaneous hemorrhage. None of the commercially prepared antihemophilic factor concentrates prepared in this country contain adequate amounts of the extremely labile bleeding time factor.

Summary in Interlingua

Le cryoprecipitato de Pool provide un efficace fonte del factor que manca in le plasma de patientes con morbo de von Willebrand e que es necessari pro un normal tempore de sanguination. Le correction del tempore de sanguination pare esser necessari pro dominar le sanguination in certe casos de sever hemorrhagia spontane. Nulle del commercialmente produciti concentratos de factor antihemophilic (disponibile in iste pais) contine adequate quantitates del extrememente labile factor del tempore de sanguination.

REFERENCES

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