An Evaluation of Crossmatching, HLA, and ABO Matching for Platelet Transfusions to Refractory Patients

By Joanna M. Heal, Neil Blumberg, and Debra Masel

Refractoriness occurs in many patients receiving multiple platelet transfusions. We used a sensitive ELISA assay to assess the utility of crossmatching HLA-A,B matched single donor platelets in 51 consecutive, typical refractory patients. Of the 222 transfusions evaluated at 1 to 4 hours posttransfusion, only 17 of 54 (31%) with positive crossmatches had corrected platelet count increments of \( \geq 7,500/\mu L \). In contrast, 95 of 168 (57%) of those with negative crossmatches had such increments \((P < .001)\). Regardless of the results of the crossmatch, HLA-A,B, and ABO matching had independent influences on transfusion outcome. The median corrected 1- to 4-hour increment for crossmatch negative transfusions was 13,300/\mu L for A/BU grade matches, 9,700 for BX, and 7,800 for C. Increments were 10,000/\mu L for ABO-identical transfusions and 5,900 for transfusions of platelets ABO incompatible with the recipient’s plasma antibodies. When the donor platelets were ABO compatible, but the donor plasma contained ABO antibodies to the recipient’s platelets, the increment was intermediate (8,200/\mu L). The most important factor in predicting platelet survival was the crossmatch, followed by HLA-A,B and ABO, each having independent predictive value. These data demonstrate that the predictive value of a negative crossmatch may be considerably less than that reported in previous studies with stable, less ill patients. In typical refractory patients, there appear to be mechanisms of platelet destruction that are related to HLA-A,B and ABO but are not detected with current crossmatch methods. We hypothesize that soluble plasma HLA-A,B and ABO antigens contribute to the destruction of donor and sometimes recipient platelets by an immune complex or other ‘innocent bystander’ mechanism. With our crossmatching technique, HLA-A,B and ABO match grades remain relevant to platelet transfusion therapy in some refractory patients.

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METHODS

Patient selection. Single donor platelets collected from a closely HLA-A,B matched, unrelated individual were crossmatched retrospectively against recipient plasma in 51 consecutive refractory patients for 316 transfusions. All patients had received RBC products from which leukocytes had not been removed, as well as numerous random donor platelet transfusions. All patients had had at least two previous transfusions of pooled random donor platelet concentrates that failed to yield satisfactory corrected platelet count increments (\( \geq 7,500/\mu L \) at 1 to 4 hours). Lymphocytotoxicity screening for anti–HLA-A,B antibodies was not routinely performed, but all patients had detectable antplatelet antibody by our enzyme-linked immunosorbent assay (ELISA) technique. Only one...
patient received granulocyte transfusions, concomitant with the platelet transfusions being evaluated by our crossmatch technique. None of these patients was free of the "nonimmunologic factors" known to shorten platelet survival. That is, all had one or more of the following factors: fever >99°F, sepsis or systemic infection, splenomegaly, consumption coagulopathy, bleeding, or a previous history of autoimmune thrombocytopenia. Determination of these factors was made by the physicians caring for the patient and was documented in the medical record. The presence or absence of these factors was not determined for each individual platelet transfusion.

The primary diagnoses were acute nonlymphocytic leukemia (21), solid tumor (5), acute lymphocytic leukemia (4), chronic myelogenous leukemia (3), preleukemia (3), aplastic anemia (2), lymphoma (2), and other hematological conditions such as myelofibrosis, erythroleukemia, multiple myeloma, etc. (11). Three of the patients were children aged <5 years. The other patients were adults. Three patients had undergone autologous or allogeneic bone marrow transplantation.

The HLA-A,B match grades of the platelet transfusions were A (7), BU (37), BX (71), and C (201). The data on the A and BU transfusions were pooled for analysis. Of the 51 patients, 25 received three or more evaluable transfusions. Of these, 16 received both B and C matches. Posttransfusion platelet count increments were measured in all patients, and the counts were corrected for blood surface area and number of platelets transfused.9 Not all patients had increments measured at both 1 and 4 to 18 hours as planned; 222 transfusions had posttransfusion platelet counts done at 1 to 4 hours. The other 94 had the initial count done between 5 and 24 hours posttransfusion; 107 transfusions had counts performed at two posttransfusion time points.

Crossmatch technique. Immulon II microtiter plates (Dynatech, Chantilly, VA) are washed three times using distilled water. A murine monoclonal antibody that reacts with an undefined antigen present on all human platelets (UR 1-6.6 produced by the Laboratory Medicine Division Hybridoma Facility) is added to each well of the microtiter plate at a concentration of -10 μg/ml in coating buffer (1.59 g/L Na2CO3, 2.93 g/L NaHCO3, pH 9.6). The plate is then incubated for 2 hours at 37°C and then overnight at 4°C. The antibody is used as crude mouse ascitic fluid. Plates are first sedimented and resuspended in phosphate-buffered saline (PBS)-EDTA (0.01 mol/L of Na2HPO4/NaH2PO4, 0.15 NaCl, 3 g/L Na2EDTA, pH 7.0) prior to adhering to the plate. After resuspension in PBS-EDTA at a concentration of 370,000/μL, a 100-μL aliquot of platelets is added to each of two wells for each plasma to be tested. After platelets are added, the microtiter plate is centrifuged at 1,800 rpm in a GLC-4 centrifuge for 10 minutes to sediment the platelets onto the monoclonal antibody-coated surface. The plate is then incubated at room temperature for 50 minutes and is washed semiautomatically, without need for centrifugation, three times using PBS (pH 7.2) and a Dynatech Mini-Wash. PBS-1% bovine serum albumin (BSA; BCA, West Chester, PA) 100 μL is added to each well to reduce nonspecific adsorption of plasma proteins and enzyme-antiglobulin conjugate during subsequent steps. This step requires a 30-minute room temperature incubation, and is followed by two further washes with PBS-1% BSA.

Test plasmas are diluted 1:2 in PBS-1% BSA to reduce nonspecific binding. Each diluted plasma (100 μL of each) is added to each of two wells and incubated for 45 minutes at 37°C if IgG is to be measured, or for 60 minutes at room temperature if IgM is to be measured. Appropriate positive (anti-Pl(A)) and negative plasma controls are also run in duplicate. The plate is washed three times using PBS-1% BSA, and 100 μL of alkaline phosphatase linked to affinity-purified anti-human IgG or IgM (Sigma Chemical, St Louis) is then added at a dilution previously optimized to the lot in question. The conjugates are usually used at 1:800 or 1:1,000 dilutions. Two wells receive no conjugate to serve as negative controls for nonspecific binding by the anti-IgG-enzyme conjugate. A further incubation at 37°C for 45 minutes (IgG) or room temperature for 60 minutes (IgM) is performed, followed by three PBS-1% BSA washes. Substrate, 1 mg/mL of para-nitrophenyl phosphate (Sigma) in 0.05 mol/L of Na2CO3, 0.001 mol/L of MgCl2, pH 9.8 is added at 100 μL/well. Color development is measured spectrophotometrically10 with a Dynatech MR600 microtiter plate reader coupled to an Apple IIE microcomputer for which kinetic-ELISA (k-ELISA) programs have been written by Dr Jonathan Cowles, Pittsford, NY: ΔA405 nm/min × 1,000 is computed by least-squares linear regression of absorbance readings taken at 20, 25, 30, and 35 minutes after substrate addition. The correlation coefficient for the linearity of the ΔA405 nm × time is usually 0.99, thus confirming the measurement of first-order enzyme kinetics, which we previously showed to correlate linearly with the amount of IgG and anti-IgG bound. The assay is linear throughout the range used.11 The amount of IgG bound to the solid-phase platelet layer is reported as the mean of the duplicate determinations of ΔA405 nm/min × 1,000. The duplicate values are usually within 10% to 20%. Because the kinetic measurement of enzyme activity is made at ambient temperature, controls are run to normalize the results from day to day.

The binding to platelets of IgG from plasmas of nonthrombocytopenic hospitalized patients yields a normal range of 0 to 1.08 for ΔA405 nm/min × 1,000 (mean ± 2 SD, n = 225). The range for binding of IgM is 0 to 9.69 (mean ± SD, n = 122). The positive control for the IgG assay is an anti-Pl(A) whose ΔA405 nm/min × 1,000 is usually ~8.00 to 10.0 when run at a 1:1,000 dilution. The positive control for the IgM assay is a plasma from a patient with leukemia and autoimmune thrombocytopenia, which usually gives a ΔA405 nm/min × 1,000 of 15 to 20 when tested at a dilution of 1:4. This latter plasma reacted with every donor platelet we tested. A positive crossmatch in both IgG and IgM assays was defined as binding >2 SD above the control means described above.

Titers of four randomly selected anti–HLA-A,B antibodies were 1 to 2 orders of magnitude higher with this k-ELISA technique than by lymphocytotoxicity assays. Titers of two anti-Pl(A) antibodies were 1 to 3 orders of magnitude greater by k-ELISA than by complement fixation techniques or 51Cr release platelet cytotoxicity methods. Thus, we believe that the technique is probably of similar sensitivity to radioimmuno or immunofluorescent methods previously shown to be useful as platelet crossmatches. Although anti-A and -B blood group antibodies are detected by this assay, many patients nonetheless exhibit negative crossmatch results with ABO-incompatible platelets.

All patient plasmas were routinely tested (crossmatched) against four to six randomly selected normal donor platelets at the same time that the crossmatch was performed with the HLA-A,B matched platelets that were transfused. All patients had at least one positive crossmatch during the course of these tests, with almost all showing incompatibility with several non–HLA-A,B matched platelets. All HLA-A,B matched platelets for transfusion were also crossmatched with two to four normal plasmas. This approach allowed us to detect donor platelets that might have significantly elevated levels of IgG or IgM. Such platelets yield positive crossmatches even with normal donor plasmas. Donor platelets with elevated platelet-associated IgG (PAIgG) or IgM occurred very rarely and were excluded from the data reported.

Platelets were stored no more than 1 to 5 days at 4°C prior to crossmatching. Such storage does not alter the level of platelet surface IgG or IgM, or the binding of plasma IgG or IgM during a crossmatch (data not shown).

Measurement of PAIgG. PAIgG assay is a technical variant of that described for the detection of plasma anti-platelet antibody.13

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results

Of the 316 IgG crossmatches performed, 88 (28%) were positive. The frequency of positive crossmatches did not vary significantly with HLA match grade: A (14%), BU (28%), BX (28%), and C (30%). This conclusion applies only to BU, BX, and C matches because only seven A matches were studied.

The IgG crossmatch positivity rate was 33 of 129 for ABO identical transfusions (26%), 26 of 90 for ABO plasma-incompatible transfusions (29%), and 31 of 97 for donor ABO platelets-incompatible transfusions (32%). However, A donor platelets crossmatched with O recipients had the highest crossmatch positivity rate—23 of 54 (43%; \( P < .02 \) v ABO identical and \( P < .07 \) v ABO plasma-incompatible transfusions).

The IgM crossmatch positivity rate did not vary with HLA or ABO match grade. Of 308 IgM crossmatches performed, 41 (13%) were positive. However, in only 15 instances were the IgM crossmatches positive and the IgG negative. There was no clear correlation between posttransfusion corrected increment and crossmatch results in these few discrepant cases.

The success rate at 1 to 4 hours posttransfusion (a corrected platelet count increment \( \geq 7,500/\mu L \)) was 95 of 168 for crossmatch-negative transfusions (57%) and 18 of 54 for crossmatch-positive transfusions (33%) \( (P < .001) \). The success rate varied significantly with HLA and ABO match grade even for crossmatch-negative transfusions: A/BU (74%); BX (62%); C (51%)—ABO identical (66%); ABO plasma incompatible (54%); ABO platelets incompatible (43%). These differences were significant for A/BU v C \( (P = 0.03) \), and for ABO identical v ABO platelets incompatible \( (P = 0.01) \).

The median corrected platelet count increments arranged according to HLA match grade for crossmatch-negative transfusions are shown in Fig 1. A similar trend was observed for crossmatch-positive transfusions, but this was not statistically significant. Corresponding data arranged according to the degree of ABO match are shown for crossmatch-negative (Fig 2) and crossmatch-positive (Fig 3) transfusions. Regardless of the crossmatch results, ABO identical platelets gave significantly better results.

The role of HLA match grade was particularly notable when time periods \( \geq 5 \) hours posttransfusion were examined (Fig 4). At these later time points, crossmatch-negative C match transfusions yielded increments in platelet count that were significantly inferior to those of A/BU and BX matches. The degree of ABO compatibility did not correlate significantly with posttransfusion increments at \( \geq 5 \) hours posttransfusion, although a trend toward better results with ABO-identical platelets was observed.

To assess the relative roles of crossmatching, HLA matching, and ABO matching in predicting platelet count increments, two- and three-way ANOVAs were performed to assess the independent and interactional contributions of these factors. Crossmatching was the most significant predictive factor for corrected platelet count increments performed 1 to 4 hours posttransfusion \( (P = .002 \) v HLA-A,B and ABO). HLA matching \( (P = .02 \) v ABO) and ABO
matching (P = .08 v crossmatching) had independent effects to a lesser degree. None of the interaction terms in these analyses was significant, confirming the largely independent roles of these three factors.

When the transfusions are grouped by crossmatch results, HLA-A,B match grade and ABO match grade, and then ranked by median corrected platelet count increment performed 1 to 4 hours posttransfusion, the independent and significant roles of these factors are clear (Table 1). Favorable HLA and ABO match grades can apparently compensate for crossmatch incompatibility in some instances. For example, the small number of crossmatch-positive A, BU and BX transfusions that were also ABO identical did as well or better than crossmatch-negative C matches that were ABO incompatible. Similarly, ABO matching can compensate for poorer HLA-A,B matching. Crossmatch-negative C matches that were ABO identical did as well or better than crossmatch-negative BX matches that were ABO nonidentical.

An unexpected finding was that after crossmatch-positive transfusions, the platelet count at 1 to 4 hours posttransfusion was frequently lower than the pretransfusion platelet count. Fifteen of 54 crossmatch-positive transfusions (27%) led to platelet count decrements as compared with only 14 of 168 (8%) for crossmatch-negative transfusions (P = .0005). A separate analysis of this phenomenon will be reported elsewhere.

To assess whether the decreased survival of transfused platelets in patients receiving crossmatch-negative transfusions might have an immunological basis, we measured PAIgG in 18 of our refractory patients at various times during the period when transfusions were given. All the refractory patients, including those receiving predominantly crossmatch-negative platelets, had elevated PAIgG at the levels seen in autoimmune thrombocytopenia (Fig 5). However, none of these patients had received exclusively crossmatch-negative, HLA-A,B-identical, and ABO-identical platelet transfusions. Elevations of PAIgG were noted after transfusions of both crossmatch-positive and crossmatch-negative platelets, as well as prior to transfusion with HLA-A,B-matched platelets.

**DISCUSSION**

Our study represents the first attempt to evaluate in one group of patients the three known immunologic factors that affect the success of platelet transfusions to refractory patients-crossmatching, HLA-A,B matching, and ABO...
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Crossmatch-positive or C-match or ABO-platelet incompatible groups fell with the BU matches. Because so few BU and BX matches fell below, of the BU or BX match groups, seven fell at or above the median category. Only two crossmatch negative groups, seven fell at or above the median, only two.

Crossmatch negative transfusions yielded unsatisfactory increments. These lower success rates in typical refractory transfusions prior to testing. Some had also recently received predominately crossmatch negative transfusions. These levels were generally measured at the nadir of the patient’s platelet count, although all patients had received multiple RBC and platelet transfusions prior to testing. Some had also recently received pooled random donor platelets. Approximately 5% to 10% of nonthrombocytopenic hospitalized patients have elevated levels of PAIgG, predominately in the range of 10 to 20 fg/platelet.

Overall failure rates reported in studies in which only stable, selected patients were reported: 8%, 20%, 8%, and 13%. Thus, nonimmunologic factors unrelated to crossmatch, HLA-A,B and ABO may account for at most ~10% to 20% of transfusion failures in typical refractory patients. An exact estimate of the role of nonimmunologic factors in platelet transfusion failures is not possible from our data because we did not examine in detail the clinical factors that might cause unsatisfactory increments in crossmatch-negative transfusions.

Our crossmatch technique appears to be sensitive to HLA-A,B, ABO and platelet-specific antibodies such as anti-PV. Therefore, it is surprising that for crossmatch-negative transfusions, HLA-A,B and ABO matching still played a significant role in predicting transfusion success. Negative crossmatches led to corrected increments of ~7,500/μL at 1 to 4 hours posttransfusion in only 57% of cases. HLA-A,B matching alone yielded a 50% success rate. The simplest possible explanation would be that our crossmatch technique is not sufficiently sensitive to HLA-A,B antibodies. We believe that this is unlikely based on its ability to detect increases in platelet-bound IgG at the level of 50 to 100 IgG molecules per platelet, and our success in detecting HLA-A,B antibodies detected by lymphocytotoxicity techniques. However, we have not made an exhaustive study of this aspect of our assay. If our platelet crossmatch assay is insensitive to some HLA-A,B antibodies, this is also likely to be true of other published assays. Other laboratories using different methodologies but similar patient selection criteria have also found low success rates for crossmatch-negative HLA-A,B matched transfusions: 52% to 77% (Table 2).

It is possible that clinically significant types of

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**Table 1. Corrected Platelet CI According to Crossmatch Results, HLA-A,B and ABO Match Grades: Ranked by Median Corrected Increment**

<table>
<thead>
<tr>
<th>CI</th>
<th>X-M</th>
<th>HLA*</th>
<th>ABO</th>
<th>n</th>
<th>Success Rate (% CI = 7,500/μL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>17,700</td>
<td>Neg BU</td>
<td>Platelets incompatible</td>
<td>7</td>
<td>86</td>
<td></td>
</tr>
<tr>
<td>17,000</td>
<td>Neg BU</td>
<td>Plasma incompatible</td>
<td>7</td>
<td>71</td>
<td></td>
</tr>
<tr>
<td>11,400</td>
<td>Neg BX</td>
<td>Identical</td>
<td>22</td>
<td>73</td>
<td></td>
</tr>
<tr>
<td>11,300</td>
<td>Neg BU</td>
<td>Identical</td>
<td>9</td>
<td>67</td>
<td></td>
</tr>
<tr>
<td>11,100</td>
<td>Pos BU/BX</td>
<td>Identical</td>
<td>9</td>
<td>56</td>
<td></td>
</tr>
<tr>
<td>10,400</td>
<td>Neg BX</td>
<td>Plasma incompatible</td>
<td>11</td>
<td>82</td>
<td></td>
</tr>
<tr>
<td>9,300</td>
<td>Neg C</td>
<td>Identical</td>
<td>43</td>
<td>63</td>
<td></td>
</tr>
<tr>
<td>5,100</td>
<td>Neg BX</td>
<td>Platelets incompatible</td>
<td>9</td>
<td>11</td>
<td></td>
</tr>
<tr>
<td>5,100</td>
<td>Pos C</td>
<td>Identical</td>
<td>10</td>
<td>40</td>
<td></td>
</tr>
<tr>
<td>4,500</td>
<td>Neg C</td>
<td>Platelets incompatible</td>
<td>26</td>
<td>42</td>
<td></td>
</tr>
<tr>
<td>4,300</td>
<td>Neg C</td>
<td>Plasma incompatible</td>
<td>34</td>
<td>41</td>
<td></td>
</tr>
<tr>
<td>3,300</td>
<td>Pos BU/BX</td>
<td>Plasma incompatible</td>
<td>2</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>1,000</td>
<td>Pos C</td>
<td>Platelets incompatible</td>
<td>14</td>
<td>29</td>
<td></td>
</tr>
<tr>
<td>900</td>
<td>Pos C</td>
<td>Plasma incompatible</td>
<td>13</td>
<td>23</td>
<td></td>
</tr>
<tr>
<td>200</td>
<td>Pos BU/BX</td>
<td>Platelets incompatible</td>
<td>6</td>
<td>17</td>
<td></td>
</tr>
</tbody>
</table>

*Only four A matches were included in these data and are grouped with the BU matches. Because so few BU and BX matches were crossmatch positive, these data were pooled for positive crossmatches.

NOTE. The median category is an increment of 5,100/μL. Of the crossmatch negative groups, seven fell at or above the median, only two fell below. Of the BU or BX match groups, seven fell at or above the median category, only two fell below. Of the five ABO-identical groups, all five fell at or above the median, none fell below. Conversely, most of the crossmatch-positive or C-match or ABO-platelet incompatible groups fell below the median group.

Abbreviations: CI, count increment.

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"Nonimmunologic" factors such as infection, consumption coagulopathy, fever, bleeding, and splenomegaly may have accounted for the fact that 43% of our crossmatch negative transfusions yielded unsatisfactory increments. Inconsistent with this interpretation is the observation that the 45 crossmatch-negative transfusions of BU or BX match grade yielded a failure rate of only 26% (top four lines in Table 1). This failure rate of crossmatch-negative, closely HLA-A,B matched transfusions compares well with the overall failure rates reported in studies in which only stable, selected patients were reported: 8%, 20%, 8%, and 13%.

Fig 5. The platelet-associated IgG (PAIgG) in 18 refractory patients, measured by a competitive enzyme-linked immunosorbent assay (ELISA) technique is shown. The amounts are in femtograms (fg) of soluble IgG equivalents per platelet.

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Anti-platelet antibody are not readily detected by any of the existing techniques, including our own. Further study of this possibility is needed before definitive conclusions can be drawn about the role of crossmatching in refractory patients.

Transfusions of donor plasma containing ABO antibodies incompatible with the recipient’s ABO antigens were associated with poorer platelet count increments in our study. Even in crossmatch-positive transfusions, HLA-A,B and ABO matching appeared to contribute to predicting post-transfusion platelet count increments. These findings cannot be explained by the insensitivity of the crossmatch used. Thus, the relatively disappointing performance of platelet crossmatching in our study and some of those previously reported are not due solely to nonimmunologic factors affecting platelet destruction or to the insensitivity of the crossmatch technique used. We suspect that the “false-negative crossmatches” are due to multiple factors in typical refractory patients.

One factor in a few instances might have been the dosage of platelets administered. A standard single donor platelet transfusion may be expected to raise the platelet count by 25,000 to 60,000/μL in a patient who is thrombocytopenic and without other reasons for decreased platelet recovery and survival. This corresponds to a corrected increment of 10,000 to 20,000/μL in a 1.7-kg adult given 4 x 10¹¹ platelets. Thus, if platelet consumption is as little as 1.3 to 3 times normal due to splenomegaly, infection, bleeding, consumption coagulopathy, or a combination of these, the corrected increment might be ≤7,500/μL solely on the basis of inadequate dosage. Unfortunately, increasing the dosage of HLA-A,B matched single-donor platelets is both logistically difficult and extremely expensive. The importance of dosage is supported by our observation that 6 of the 15 satisfactory increments observed despite positive platelet crossmatches occurred in very small children receiving full adult dosages of platelets. Baldini and colleagues reported that giving very large dosages of incompatible platelets to children unresponsive to platelet transfusion could temporarily overcome their refractoriness.

A second possible explanation for false-negative crossmatches is that immunological causes of refractoriness exist other than antibody specific for platelet surface antigens. If this is so, it should not be surprising that in vitro assays such as crossmatching fail to predict in vivo results consistently. Indeed, our data show that in vivo findings, such as post-transfusion platelet count increments and level of IgG on circulating platelets, are not fully predicted by the results of crossmatching. Thus, additional hypotheses for immunologic mechanisms of platelet destruction in typical refractory patients are in order.

The additional causes of platelet destruction in refractory patients, not detectable by current crossmatch techniques, are probably immunologic. We base this conclusion on the important role that HLA-A,B matching and ABO matching appear to play in transfusions without detectable antiplatelet antibody. Also, our refractory patients universally had platelets coated with large quantities of IgG. This increased IgG may be due to previous incompatible platelet transfusions, as none of our patients received only crossmatch-negative, HLA-identical, and ABO-identical transfusions. Elevations in PAIgG were noted prior to transfusion and after both crossmatch-positive and crossmatch-negative transfusions. Thus, this IgG may be a nonspecific finding of no clinical significance. This increased IgG may also be due to immunologic mechanisms other than platelet-specific antibody...
detectable in a crossmatch. Such immunologic mechanisms could be characteristic of the refractory state.

Two possibly relevant mechanisms are suggested by previous investigations of immune destruction of platelets due to drugs and microorganisms. Shulman and Jordan23 and, more recently, Christie and Aster24 demonstrated that platelets can acquire drug and IgG complexes by at least two pathways. One is the binding of IgG-drug complexes to platelet Fc receptors. The other is the binding of IgG, drug, and platelet in a complex that requires all three for antibody fixation on the platelet. Kelton and associates showed that the thrombocytopenia of malaria is due to acquisition of malarial antigens and the corresponding IgG antibody by the patient’s platelets.25 We propose that similar innocent bystander mechanisms involving soluble donor plasma HLA-A,B and ABO antigens may exist. This hypothesis is supported by our finding that, independent of the crossmatch result, HLA-A,B and ABO matching predict the posttransfusion platelet count increment. Platelets can acquire new blood group antigens in the anti-A from the donor plasma combines with soluble A antigen in the recipient’s plasma, and then damages the type O donor platelets by an immune-complex mechanism. Alternatively, the type O transfused platelets may acquire A antigen in the recipient’s plasma and subsequently be injured directly by the anti-A from the donor. If such mechanisms exist, incompatible transfusions might affect the fate of subsequent transfusions that were otherwise immunologically compatible. Any role for plasma HLA-A,B and ABO antigens in platelet transfusion immunology remains speculative and will require further investigation.

If our preliminary results are confirmed by other researchers, the role of platelet crossmatching in typical refractory patients clearly may be more limited than has been expected. We and others had hoped that crossmatching would make HLA-A,B matching unnecessary and enable the use of randomly selected crossmatch-negative pools of donor platelets. Our data do not provide much cause for optimism in this regard, at least in typical refractory patients. In our patients, crossmatch-negative C-matched platelets do reasonably well in the first few hours posttransfusion. However, later C-matched platelets appear to be considerably less likely to provide satisfactory platelet count increments than do A-, B-, or BX-matched platelets. Some of this disadvantage may be overcome by using ABO-identical C-matched platelets. Our findings support a role for ABO in platelet transfusions, first studied by Aster ~20 years ago.30 New data on possible roles for ABO31 and HLA-A,B32 matching have recently been reported by other investigators.

Our study has certain practical implications for changing the manner in which platelet donors are selected for refractory patients. If an IgG crossmatch technique is available and practical, it should be used to avoid transfusing donor platelets that are crossmatch positive with the recipient. Closely HLA-A,B-matched platelets should be given next priority. If C or worse grades of match are the only ones available, ABO identical donors should be selected if feasible. Our data suggest that an IgM crossmatch will not add significantly to the clinical benefit to be gained by crossmatching in most cases.

The alleviation of refractoriness in many patients may be quite difficult, if our results are typical. Further research into the multiple mechanisms of immunologic refractoriness to platelet transfusion is indicated, and efforts to prevent refractoriness warrant additional attention.

NOTE ADDED IN PROOF

Brand et al recently reported ABO antibodies as a cause of platelet refractoriness (Transfusion 26:463, 1986).

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An evaluation of crossmatching, HLA, and ABO matching for platelet transfusions to refractory patients

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