Effect of Intentional Hemodilution on Platelet Survival in Secondary Pulmonary Hypertension

Antonio A.B. Lopes, M.D.;† Nair Y. Maeda, M.S.; Munir Ebaid, M.D.;‡ Dalton F. Chamone, M.D., Ph.D.§; and Fulvio Pileggi, M.D.¶

Platelet regeneration time was assessed in 13 young adults with pulmonary hypertension and polycythemia secondary to congenital heart defects who underwent isovolemic hemodilution to improve clinical status and coagulation defects. The estimated platelet half-life in patients with Eisenmenger's complex was significantly shortened in comparison with normal subjects (3.8±1.9 vs 4.8±1.0 days, p<0.05). Hemodilution was carried out with no adverse effects, using low molecular weight dextran solutions. Lowering hematocrit from 61 to 50 percent resulted in a significant increase in platelet half-life from 3.8±1.9 to 5.7±1.8 days (p<0.02), which was followed by a marked rise in platelet count from 149±31 to 209±47×10⁶ platelets/L (p<0.003). Arterial oxygen tension did not change significantly. These observations indicate that high hematocrit levels may have accounted for the shortened platelet survival and thrombocytopenia in these patients. Significant hemodilution may lead to a marked improvement in platelet abnormalities in patients with Eisenmenger's complex.

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It is now generally accepted that platelets and coagulation play an important role in the pathophysiologic mechanisms of pulmonary hypertension. Hypoxia and polycythemia may occur in patients with pulmonary vascular disease secondary to congenital heart defects and may lead to additional changes in coagulation and platelet function. These abnormalities also contribute to further enhancement of pulmonary vascular reactivity and increase in pulmonary resistance.

Many authors, such as Rosenthal et al. and Wedemeyer and Lewis have demonstrated improvement in hemostasis following red cell volume reduction in cyanotic patients with congenital heart defects. However, in these studies, patients with and without pulmonary hypertension were included. The aim of this report is to evaluate the platelet survival in a specific group of young adults with Eisenmenger's complex and to investigate the extent it can be improved by intentional hemodilution, lowering hematocrit to a predetermined level.

PATIENTS AND METHODS

We studied 13 patients, ages 17 to 40 years (median 27 years), with congenital heart defects, pulmonary hypertension and polycythemia (Eisenmenger's complex), who underwent intentional hemodilution to improve the clinical status and coagulation abnormalities. The associated cardiac defects were: ventricular septal defect (six patients), univentricular heart (three patients), atrial septal defect (two patients) and double outlet right ventricle (two patients). The mean pulmonary arterial pressure in this group ranged from 52 to 104 mm Hg, and before hemodilution, all of them were in NYHA functional class 2, 3 or 4.

The hematocrit (Hct) ranged from 54 to 67 percent and the hemodilution procedure was planned to achieve a final Hct of 50 percent. The calculation of the blood volume to be extracted was based on a previously reported logarithmic model:

\[ V = V_0 \log (Hct) - \log (Hct_r) \]

where:

- \( V \) = circulating blood volume
- \( V_0 \) = initial and final values

Extracted blood was simultaneously replaced by an equal volume of low-molecular weight dextran (LMWD, MW 40,000) and the velocity of exchange did not exceed 10 ml/min. Arterial oxygen tension (PaO₂) was determined before and after hemodilution.

Platelet survival and count were assessed in all patients before and one week after the hemodilution procedure, to avoid interference of dextran on platelet function. Platelet counting was evaluated by the direct Brecher-Cronkite method using phase microscopy. Platelet survival was assessed by the nonisotopic method based on regeneration of malondialdehyde (MDA) after intake of acetylsalicylic acid (ASA), which irreversibly blocks platelet MDA production by inhibiting cyclooxygenase. Therefore, the rate of platelet MDA regeneration represents the appearance of new platelets in the blood stream. Daily measurements of platelet MDA content were performed for ten days after intake of a single dose of 500 mg ASA. For each patient, MDA measurements were always reported as percentage of baseline value for 10⁴ platelets. In steady-state conditions, the time necessary for the regeneration of 50 percent of baseline platelet MDA content was considered the estimation of platelet half-life (T½). For the determination of T½, MDA production was measured in platelets using a heat-induced reaction with 2-thiobarbituric acid to obtain a pink colored substance. The optical density was read at 532 nm in a spectrophotometer. Patients with less than 300×10⁶ platelets/L in the assay were excluded, to avoid
Table 1—Effect of Hemodilution on Platelet Count and Survival

<table>
<thead>
<tr>
<th></th>
<th>Hct (%)</th>
<th>T_{wa} (days)</th>
<th>Platelets (×10^9/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal control subjects (N = 20)</td>
<td>47 ± 2</td>
<td>4.8 ± 1.0</td>
<td>242 ± 61</td>
</tr>
<tr>
<td>Eisenmenger's complex (N = 13)</td>
<td>61 ± 3</td>
<td>3.8 ± 1.9↑</td>
<td>149 ± 31↑</td>
</tr>
<tr>
<td>baseline after hemodilution</td>
<td>50 ± 2*</td>
<td>5.7 ± 1.8§</td>
<td>209 ± 47§</td>
</tr>
</tbody>
</table>

Mean ± SD.  
†p<0.05; ‡p<0.003 vs normal control subjects  
*p<0.002; ‡p<0.003 vs baseline

Statistical Analysis

Changes in hematocrit, platelet count and platelet MDA content, were analyzed using the Wilcoxon signed-rank test. We have previously demonstrated that parametric tests may be used in studies of T_{wa}. For each patient, T_{wa} was calculated using linear or logarithmic regression analysis according to the best coefficient of determination. Mean T_{wa} values in normal subjects and in patients with Eisenmenger's complex before and after hemodilution were compared using Student's t-test for paired and unpaired observations. Two-tailed tests with significance level of 0.05 were used.

The results are reported as mean ± SD.

RESULTS

Basal, post-hemodilution and reference values of Hct, platelet count and T_{wa} are shown in Table 1. The lines of platelet MDA regeneration for each group of observations are represented in Figure 1 as confidence intervals. Platelet count in patients with Eisenmenger's complex and Hct level of 61 percent was decreased when compared with normal subjects (p<0.003). The displacement of MDA regeneration line to the left indicated that the platelet survival was shortened (Fig 1).

Hemodilution was carried out with no adverse effects and it was followed by clinical improvement in seven of nine patients in NYHA class 3 or 4. A slight rise in the mean PaO_2 value from 43.5±5.3 to 45.5±6.2 mm Hg was observed. The hematocrit significantly decreased from 61±3 to 50±2 percent (p<0.002), and this resulted in a marked improvement in platelet half-life from 3.8±1.9 to 5.7±1.8 days (p<0.02). The improvement in platelet survival, which is indicated by the shift in MDA regeneration line to the right (Fig 1), was followed by a significant increase in platelet count from 149±31 to 209±47×10^9 platelets/L (p<0.003). In addition, hemodilution resulted in a significant rise in pre-ASA platelet MDA content from 2.03±0.86 to 3.62±1.12 nM/10^9 platelets (p<0.002).

DISCUSSION

Previous studies have demonstrated that coagulation abnormalities and platelet disorders may occur in patients with congenital heart defects and polycythemia, especially children with and without pulmonary hypertension. In the study of Wedemeyer and Lewis, multiple phlebotomies have proved beneficial in improving these hemostatic defects. The volume of blood removed was calculated on the basis of body surface area, and as a result, the final Hct varied considerably from patient to patient.

Specific studies of platelet function have been carried out in patients with hypoxemia and pulmonary hypertension and many have revealed decreased platelet count and survival. In our study we evaluated the platelet survival in a specific group of 13 young adults with Eisenmenger's complex and we examined the extent it could be changed by lowering Hct to a predetermined level of 50 percent. It has been reported that the fall in oxygen capacity of the blood is fully compensated by flow increase. Hyperoncotic LMWD solution was used for this purpose, considering its beneficial effects on blood rheology. Although it has been shown that phlebotomies can be

![Figure 1](http://journal.publications.chestnet.org/pdfaccess.ashx?url=/data/journals/chest/21595/ on 04/28/2017)
safely performed without fluid replacement in cyanotic patients,3 blood exchange for dextran was the procedure of choice in this study, since large amounts of blood were removed to achieve the final Hct of 50 percent. Functional improvement was observed after hemodilution in seven of nine patients in previous class 3 or 4. The time necessary for the Hct to return to baseline level ranged from three to four months in our patients.

Controversy exists about the accuracy of platelet survival estimation by the nonisotopic MDA method.16 The main difficulties concern platelet MDA measurements in extremely thrombocytopenic subjects. For this reason we excluded patients in whom a minimum of $300 \times 10^9$ platelets/L could not be obtained for assay. Reference normal values of platelet half-life observed in the present study are in accordance with previous reports.8,14,20 It is of interest that the method used to determine platelet half-life might theoretically change that half-life, since aspirin may alter thromboxane-prostacyclin equilibrium. However, there is no available information that a single dose of aspirin can significantly change platelet survival in chronic hypercoagulable states. Furthermore, it is likely that the influences of the single dose of aspirin on platelet metabolism and thromboxane-prostacyclin equilibrium were similar, before and after hemodilution. Thus, the change in platelet half-life observed in our patients does not seem to be related to aspirin intake.

Our data indicate that platelet survival is shortened in Eisenmenger’s complex. Very similar decrease in platelet life-span has been demonstrated in pulmonary hypertension and hypoxia secondary to chronic obstructive pulmonary disease.3,4,11 The improvement in platelet survival in our patients was followed by a significant rise in platelet count.

The reason why platelet count and survival are decreased in Eisenmenger’s complex is not fully understood. Intravascular coagulation has been presented as a possible cause of platelet consumption in cyanotic patients.1,2,10 Platelet aggregation in the pulmonary microcirculation would significantly contribute to further enhancement of vascular resistance by the release of vasoactive substances and growth factors.21,22 We speculate that the improvement in coagulation abnormalities after hemodilution may have accounted for the increase in platelet life-span in our patients. The lower level of platelet MDA content observed before hemodilution may represent chronic in vivo platelet activation, followed by release reaction. Indeed, these “tired” or “depleted” platelets associated with circulating platelet aggregates have been demonstrated in patients with vaso-occlusive disorders.23,24 We observed a significant increase in platelet MDA content after hemodilution and this may indicate that in this condition, platelets are involved in fewer activation reactions and secretory responses.

The role of hypoxemia as a major cause of decreased platelet count and survival in these patients remains controversial. Terai et al20 have demonstrated a direct significant relationship between capillary oxygen tension and platelet count in infants with transposition of the great arteries. However, an inverse relationship between Hct and platelet count has been shown. It is possible, therefore, that secondary polycythemia rather than hypoxemia itself represented the main cause of thrombocytopenia in their patients. In our study, the increase in platelet survival and count occurred in the absence of significant change in PaO$_2$.

Our findings strongly suggest that hyperviscosity and sludging of red blood cells associated with damaged endothelium are the major causes of shortened platelet survival in Eisenmenger’s complex. The role of platelets in the pathophysiologic mechanisms of pulmonary hypertension warrants investigation of therapeutic measures to improve platelet abnormalities. Further studies are necessary for a better understanding of the consequences of platelet-vessel interactions in this syndrome.

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