PLATELET DESTRUCTION AND SEROTONIN RELEASE DURING HAEMODIALYSIS

L. J. LAWSON, N. CRAWFORD, P. DAWSON EDWARDS and J. D. BLAINEY

The Artificial Kidney Unit, Queen Elizabeth Hospital, Birmingham, Great Britain

The circulation of blood in extra-corporeal systems inflicts a varying degree of mechanical trauma on the cellular elements. This has been well documented in studies performed during cardio-pulmonary bypass with regard to platelet and red cell destruction (Salzman, 1963). Although the blood flow rates employed in this procedure are considerably higher than those used with haemodialysis, we have observed alterations in platelet counts during dialysis which may be of metabolic significance.

The blood platelets in addition to their known role in haemostasis, also transport the potent vaso-active agent serotonin (or 5-hydroxytryptamine-5 HT), from sites of production and secretion in the gastro-intestinal tract to areas of functional release or enzymatic breakdown. The purpose of this communication is to present our findings, concerning the magnitude of platelet losses which have been observed during 30 routine haemodialyses carried out in each case for a period of six hours. In addition, eight of these patients, selected at random, were subjected to more detailed investigations in order to establish the site of platelet loss within the system. Determinations were also made of the platelet bound and free plasma serotonin levels because of the possible association between liberated platelet content and the occurrence of post-dialytic complications.

Materials and methods
All 30 patients in this study had oliguric renal failure. 19 were suffering from acute failure following surgery and 11 were in the terminal phase of chronic renal disease. Standard techniques of haemodialysis using the Kolff twincoil artificial kidney were employed with non-interrupted bath water change at two-hourly intervals. The blood flow rate varied from 250-400 ml/min. The sites from which blood samples were withdrawn are shown in Figure 1. The samples were collected in siliconised tubes contained E.D.T.A./saline as an anticoagulant. Platelet counts were made immediately, using a bulk dilution procedure and

![Diagram]

Fig. 1. Haemodialysis system indicating sites from which blood samples were withdrawn.

63
carried out in duplicate by phase contrast microscopy. Platelet bound and free plasma serotonin levels were determined by spectrophotofluorometric methods reported previously.

![Diagram](image)

**Fig. 2.** Whole Blood Platelet Counts, before and after dialysis. $\times 10^3$/cu. mm.

<table>
<thead>
<tr>
<th>CASE SAMPLE SITE</th>
<th>J.B. 1, 4</th>
<th>J.F. 1, 4</th>
<th>J.M. 1, 4</th>
<th>MEAN % FALL</th>
</tr>
</thead>
<tbody>
<tr>
<td>ONSET</td>
<td>180-159</td>
<td>167-130</td>
<td>268-201</td>
<td>-20</td>
</tr>
<tr>
<td>MID</td>
<td>130-139</td>
<td>-</td>
<td>-</td>
<td>+3</td>
</tr>
<tr>
<td>END</td>
<td>121-128</td>
<td>117-114</td>
<td>195-198</td>
<td>+2</td>
</tr>
</tbody>
</table>

**Fig. 3.** Whole Blood Platelet Counts, during dialysis, from sample sites 1 and 4 $\times 10^3$/cu. mm.

<table>
<thead>
<tr>
<th>CASE SAMPLE SITE</th>
<th>J.W. 2, 3</th>
<th>E.G. 2, 3</th>
<th>J.M. 2, 3</th>
<th>MEAN % FALL</th>
</tr>
</thead>
<tbody>
<tr>
<td>ONSET</td>
<td>141-78</td>
<td>257-208</td>
<td>256-212</td>
<td>-27</td>
</tr>
<tr>
<td>MID</td>
<td>89-85</td>
<td>213-188</td>
<td>-</td>
<td>-8</td>
</tr>
<tr>
<td>END</td>
<td>102-99</td>
<td>152-124</td>
<td>189-186</td>
<td>-8</td>
</tr>
</tbody>
</table>

**Fig. 4.** Whole Blood Platelet Counts during dialysis, from sample sites 2 and 3, $\times 10^3$/cu. mm.

* A statistical analysis of the precision of platelet counting revealed standard deviations of $4.0 \times 10^9$ per cu.mm. for 86 unselected duplicate whole blood counts, and $1.7 \times 10^9$ per cu.mm. for 226 consecutive platelet-rich plasma counts. Calculations were made by the method of duplicate difference.
PLATELET DESTRUCTION AND SEROTONIN RELEASE DURING HAEMODIALYSIS

(Crawford and Rudd, 1962, Crawford 1963, 1965). The platelet serotonin concentration is expressed as nanograms per 10^9 platelets and free plasma serotonin as nanograms per ml of plasma (one nanogram is one thousandth of a microgram).

Results

Whole blood platelet counts before and after dialysis show a mean fall of 38% as shown in Figure 2 (P. 0.001). Included also in this figure are the platelet count results for nine determinations in the eight patients in whom serotonin measurements were also made. The mean fall for this group is similar, namely 34% (P. 0.001). Figure 3 shows the results for three patients of the whole blood platelet counts on paired samples taken simultaneously from sites 1 and 4 at the onset of dialysis, at a point mid-way in the procedure, and at its termination. It will be seen that the greatest platelet loss occurs at the start of dialysis (a fall of 20% on a mean basis.) Little significant change in the platelet count occurs after this initial period. The values for the middle and the end of dialysis are within the confidence limits of platelet counting. Figure 4 shows the changes in whole blood platelet counts in three further cases from whom paired samples were taken at sites 2 and 3 in the extra-corporeal circuit at similar times during dialysis. A mean platelet loss of 27% was recorded in the onset samples and again little change occurs at later periods in the dialysis. Since these results are essentially the same as those in Fig. 3, it suggests that the coil is largely responsible for the platelet losses that are observed.

Fig. 5 compares the fall in whole blood platelet counts occurring between the start and mid-point of dialysis with the concentration of serotonin within the platelets when measured at the same time. It is seen that as the platelet count falls (mean decrease 34%) there is a reciprocal increase in the platelet serotonin concentration. A mean increase of unit concentration of 58% was observed. Little further change in whole blood platelet counts occurred during the latter half of dialysis, and the platelet serotonin concentration falls towards the

![Diagram](image)

Fig. 5. Change in whole blood platelet counts and platelet serotonin concentration during dialysis.

65
onset value. It still shows at the end of dialyses, however, an increase of 16% on a mean basis. In five of these patients parallel studies of thromboplastin regeneration showed no abnormality when compared with normal volunteer controls.

Discussion and summary

The loss of circulating platelets reported in cardiopulmonary bypass occurs usually within minutes of the onset of perfusion (Salzman, 1963). It would appear that a similar rapid response is encountered during haemodialysis and in the 30 patients studied here, an average of one third of the total circulating platelets were lost during the procedure. Paired samples taken at the onset of haemodialysis show a dramatic fall in whole blood platelet count, and this is reduced by the mid-point of a six-hour dialysis to only a 2 or 3% fall. Comparison of values taken at different sample sites suggest the cellophane coil as the principal site of platelet destruction. This is perhaps not surprising when one considers the surface area involved which is almost 2 sq. metres in the case of the Kolff twin coil pack.

When blood platelets are destroyed, free serotonin is liberated. The antidiuretic effect of this amine has been well documented in both animals and man (Ersparmer and Ottolenghi, 1953; Frick, 1960; and Bojs, 1961). Furthermore, Frick, Virkkula and Paasonen in 1961 have showed that following major surgery, platelet breakdown is associated with a reduction in urinary output. Following haemodialysis, there is frequently a transient reduction in the already low daily urine volume. In the studies reported here, the platelet losses occurring during the early phases of haemodialysis are accompanied by reciprocal increase in the serotonin concentration of the remaining circulating platelets. At the time of platelet removal the plasma free serotonin levels do not increase. It is of interest, however, that the levels of this fraction in patients with renal failure have been found to be slightly higher (mean 29 ng/ml, range 4-62 ng/ml) than our earlier findings reported for 27 normal subjects in whom a mean value of 13 ng/ml, range 0-29 ng/ml was found (Crawford, 1965). The reasons for these differences are not yet apparent. We feel, however, that it is unlikely that platelet released serotonin is related to post-dialysis anti-diuresis. Our results taken in conjunction with the normality of thromboplastin regeneration tests support the view that the remaining circulating platelets present after haemodialysis are functionally and metabolically competent both with respect to their clot forming propensity and their capacity for the removal of undesirable neurohumoral agents in the plasma.

Acknowledgement

We should like to acknowledge the receipt of a full-time grant to L. J. L. from the Endowment Fund, United Birmingham Hospitals.

REFERENCES


DISCUSSION

The CHAIRMAN: Any questions or remarks?

Dr. D. N. S. Kerr (Newcastle): How much heparin are you priming your patients and coils with?

Dr. L. J. Lawson (Birmingham): All these patients had 10,000 units of heparin within the circulating system at the start of the dialysis, and this was repeated as necessary later in the procedure, on the basis of the clotting times of the patient and the circuit.