Effects of Hemorrhagic Shock on Vascular Permeability to Red Blood Cells

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The studies of Drinker have indicated that red blood cells or extrinsic particulate material may appear in the lymph without morphological changes of the small blood vessels. More recently, Bigelow, Furth, Woods, and Storey showed that the number of red blood cells in the lymph increases during thrombocytopenia induced by x-irradiation as a result of increased permeability of the small blood and lymph vessels to red blood cells. Lymph duct cannulation techniques were subsequently applied to the study of thrombocytopenic hemorrhage and of the hemorrhagic tendency associated with the use of anticoagulants.

Spontaneous intestinal hemorrhages are frequently observed in advanced stages of shock. In the present studies, the permeability of blood vessels to red cells, as determined by changes of the output of red blood cells in lymph, was investigated in dogs in hemorrhagic shock.

Methods

Healthy mongrel dogs (10 to 16 Kg. body weight) were used. The thoracic lymph duct was cannulated under Nembutal anesthesia (45 mg./Kg.) according to the technique of Brown and Hardenbergh. Following administration of Heparin (2 mg./Kg. body weight), hemorrhagic shock was induced, as described by Fine et al. The mean blood pressure (femoral artery) was maintained at 30 mm. Hg.

In a number of experiments, the administration of heparin was repeated at intervals of three hours. The blood which had been withdrawn was reinfused when more than 40 per cent of the original volume had returned from the attached reservoir to the circulation in order to maintain the blood pressure at 30 mm. Hg or after six hours of hypotension, whichever was shorter.

Lymph was collected at regular intervals for periods of five minutes. During the intervals, the flow of the lymph from the left thoracic duct into the jugular vein was restored by connecting the corresponding plastic cannulae.

The total output of red blood cells per minute was determined by direct counting of the red blood cells and measuring the volume of lymph collected. Platelet counts were carried out under phase-contrast microscopy by the technique of Brecher and Cronkite.

Results

In control studies on five dogs, 2 mg. of Heparin per Kg. of body weight were administered intravenously every three hours. The output of red blood cells in the lymph collected from the thoracic lymph duct during periods of six and one-half to 11 hours remained normal on all occasions (i.e., $10^6$ to $10^7$ red cells per minute).

Hemorrhagic shock was induced in 10 animals. In three dogs, hypotension was maintained for periods of one to two hours only. In two animals (dogs, nos. 45 and 136 B), the lymph remained unchanged during the hypotension and following its termination by transfusion. On both occasions, the minimal blood pressure was successfully maintained at 30 mm. Hg (fig. 1). The third animal (dog no. 129), however, experienced a sudden and unexplained drop of the blood pressure to zero, which was corrected by rapid transfusion. Moderate increase of the output of red cells in the lymph was observed five hours later (table 1).

Seven additional animals were bled and kept hypotensive for a maximum of six hours, or until more than 40 per cent of the blood...
output of red blood cells in the lymph during and after hemorrhagic shock of short duration. removed had to be reinfused in order to maintain the mean arterial blood pressure at 30 mm. Hg. The output of red blood cells in all animals in this group increased markedly following the restoration of the blood pressure to normal by transfusion of the blood remaining in the reservoir (figs. 2 and 3, table 1). It should be noted that on three occasions (in dogs no. 159, 126, and 210), the lymph became moderately bloody prior to the reinfusion. One animal (dog no. 140) was rendered hypotensive on two consecutive occasions. Bleeding into the lymph occurred only after the second hypotensive phase during which more than 40 per cent of the shed blood had reentered the circulation (fig. 4).

The lymph flow was reduced during the period of hypotension in six experiments and remained relatively unchanged in one. In six of the seven experiments, the lymph flow after reinfusion of the shed blood was greater than prior to induction of hypotension. On two occasions (in dogs no. 108 and 119), the platelet counts after restoration of the blood volume were moderately decreased (—34 per cent and —36 per cent). In the majority of animals studied, however, significant changes of platelet levels were not observed.

Postmortem examination carried out eight hours following induction of hypotension (in dogs no. 119, 108, and 140) failed to reveal gross hemorrhages on the surface, in the lumen of the intestines, or on the peritoneum or the mesentery. Animals no. 159 and 126, which died after 12 and 17 hours, respectively, showed bloody peritoneal fluid and multiple petechiae and ecchymoses in the above structures.

Discussion

Hypotension in dogs, due to blood loss, was followed by increased output of red blood cells in the lymph collected from the thoracic lymph duct. In these experiments, bleeding into the lymph occurred after prolonged hypotension (six hours) or following spontaneous return ("uptake") of more than 40 per cent of the withdrawn blood. These criteria have previously been found to correlate with the induction of irreversible shock.8 Further studies are necessary, however, to determine whether the irreversibility of hemorrhagic shock is associated with a widespread vascular damage manifested also by an increased permeability of the small blood and lymph vessels to red blood cells.

The changes of lymph composition reported above are comparable to these in thrombocytopenia, characteristically associated with an increased output of red blood cells in the lymph.2 The mechanisms responsible for the increased output of red blood cells into the lymph of dogs with hemorrhagic shock are however not clear. Wide-spread thrombosis of small blood vessels has been observed in shock and was suggested to account for hemorrhages10 found in dogs dying from irreversible hemorrhagic shock. In the studies presented here, the use of Heparin in single or repeated doses reduces the likelihood of intravascular clotting. The amount of Heparin (2 mg. Kg.)
FIGURE 3
Lymph samples from a dog during and after hemorrhagic shock.

does not appear to be responsible, by itself, for the increased output of red blood cells in the lymph, as indicated by the corresponding control studies. Previous studies on rats, however, have shown that delayed bleeding into the lymph can follow the administration of relatively large amounts of heparin. Heparin may have therefore contributed to the hemorrhagic tendency described above.

The effects of the rapid changes of blood

TABLE 1
Output of Red Blood Cells in the Lymph of Dogs in Hemorrhagic Shock

<table>
<thead>
<tr>
<th>Dog no.</th>
<th>Duration of hypotension</th>
<th>Amount of blood withdrawn (cc.)</th>
<th>Amount of blood infused (cc.)</th>
<th>RBC output just prior to reinfusion $\times 10^6$</th>
<th>Maximum RBC output $\times 10^6$</th>
</tr>
</thead>
<tbody>
<tr>
<td>45</td>
<td>2 hours</td>
<td>450</td>
<td>450</td>
<td>0</td>
<td>5</td>
</tr>
<tr>
<td>136 B*</td>
<td>2</td>
<td>835</td>
<td>835</td>
<td>5</td>
<td>5</td>
</tr>
<tr>
<td>129†</td>
<td>1 1/2</td>
<td>420</td>
<td>420</td>
<td>8</td>
<td>2751</td>
</tr>
<tr>
<td>140</td>
<td>3</td>
<td>545</td>
<td>330</td>
<td>0</td>
<td>200</td>
</tr>
<tr>
<td>126</td>
<td>4</td>
<td>600</td>
<td>360</td>
<td>75</td>
<td>2,700</td>
</tr>
<tr>
<td>119</td>
<td>3</td>
<td>635</td>
<td>380</td>
<td>5</td>
<td>1,600</td>
</tr>
<tr>
<td>108</td>
<td>3</td>
<td>640</td>
<td>385</td>
<td>12</td>
<td>590</td>
</tr>
<tr>
<td>159</td>
<td>6</td>
<td>360</td>
<td>220</td>
<td>50</td>
<td>1,000</td>
</tr>
<tr>
<td>127</td>
<td>5 1/4</td>
<td>500</td>
<td>300</td>
<td>0</td>
<td>2,300</td>
</tr>
<tr>
<td>210</td>
<td>5 1/4</td>
<td>660</td>
<td>396</td>
<td>20</td>
<td>1,200</td>
</tr>
</tbody>
</table>

*Two episodes of hypotension of short duration were induced successively in this dog.
†Sudden fall of blood pressure to zero during hypotensive phase.
VASCULAR PERMEABILITY IN SHOCK

volume and of blood pressure were investigated in dogs no. 45, 136 B, and 129. Hypo-
tension of short duration in these animals had no effect on the output of red blood cells
in the lymph in dogs no. 45 and 136 B, while a slight increase occurred in dog no. 129. The
last animal, however, differed from the others by experiencing a sudden and unexplained
drop of the blood pressure to zero.

Although on two occasions some decrease of the levels of circulating platelets was ob-
erved (dogs no. 108 and 119) increased output of red blood cells occurred also without
significant changes of platelet levels.

Erythrophagocytosis could also be consid-
ered to account for the appearance of red
cells in the lymph, especially in view of the
temporary extravasation of large amounts of
blood and its storage in a glass reservoir. It
should be noted in this regard, that at least
in three (dogs no. 126, 159, and 210) of the
seven animals subjected to conditions known
to result in irreversible shock, slight but defi-
nitely abnormal increase of the output of red
blood cells in the lymph was observed prior
to transfusion of the blood initially removed.
In addition, the red cells in the lymph were
free and in good condition. It may also be
difficult to explain hematocrits of the lymph
as high as 20 per cent to 25 per cent (dogs
no. 126 and 127) on the basis of erythrophago-
cytosis alone. It is very likely, therefore, that
the bleeding into the lymph reported above
may be associated with endothelial damage
analogous, although perhaps not identical, to
this observed by Bigelow, Furth, Woods, and
Storey\(^2\) in animals exposed to x-irradiation.

The need for adequate intravascular pres-
sure for diapedesis of red cells across normal\(^1\)
or damaged by thrombocytopenia vessels (un-
published observations) probably accounts for
the delay of severe bleeding in our experi-
ments until normal or near-normal blood pres-
sure had been re-established.

The role of anemic anoxia or of changes in
blood pressure levels in the portal system,
in blood coagulation, and in the fibrinolytic
system, cannot be assessed at this time on
the basis of the observations described. The possi-

![FIGURE 4](http://circres.ahajournals.org/)

Red blood cells output in the lymph after induction of hemorrhagic shock of short and prolonged duration in the same animal.

The lymph collected from the left thoracic
lymph duct of dogs in severe shock due to
blood loss becomes grossly hemorrhagic. Re-
versible hemorrhagic shock is not accompanied
by changes in lymph appearance or content
of red blood cells. The increased levels of
red cells in the lymph was observed prior to
the appearance of the characteristic gross
intestinal and peritoneal hemorrhages.

Summary

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