The Role of the Spleen in the Leukocytosis Following the Intra-arterial Administration of Epinephrine

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One of the well-known hematologic reactions to the intravenous administration of epinephrine is a significant rise of leukocyte numbers in the peripheral blood. Since the work of Harvey in 1906 and of Frey in 1914, this leukocytosis has been associated with contraction of the spleen with resultant release of the cells, and has been extrapolated as a demonstration that the spleen is one of the major reservoirs of leukocytes in the body. The height of the leukocytosis following epinephrine has been employed as a measure of the contractility of the spleen and its sequestered cellular content. The literature dealing with this concept is controversial, confirming or denying the observations. Harvey's conclusions were based on single experiments on a dog, cat, and rabbit. Frey believed that the lymphocyte response was a functional test of the contractility of the spleen but it is too complicated to be explained simply by splenic contraction. Barcroft was unable fully to support Harvey or Frey's contention from his data despite his extensive studies of the splenic circulation in the dog. Camp found the lymphocytosis following pilocarpine administration in splenectomized patients to be as profound as when the spleen was present. Similar data were obtained in other animals. Lucia and his associates supplied the most conclusive data refuting Frey's test as a function of splenic contractility in man when they found similar leukocytoses following subcutaneous epinephrine administration in 4 patients both before and after splenectomy.

On the basis that epinephrine causes a marked increase in thoracic lymph flow, it has been concluded by some investigators that the lymph is the chief contributory, if not the sole source, of lymphocytes after exercise or pilocarpine and epinephrine injection. Contraction of the lymph nodes has also been considered as another possible origin of lymphocytosis. The liver has also gained some attention as a site of leukocyte sequestration but the studies of the reaction of the liver to epinephrine with respect to leukocyte changes in the blood has not confirmed this suspicion.

The majority of the studies on the contractile effects of epinephrine upon the spleen have been performed on animals, the dog in particular, while many of the hematologic alterations of clinical interest have been ascertained in man.

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Much extrapolation from the animal data has been employed to supplement that which has been obtained from patients, frequently resulting in controversial conclusions.

The spleen of the dog contains many smooth muscle fibers in the capsule and the trabeculae which permeate the splenic pulp and are considered ample to explain the contraction of the spleen following the administration of epinephrine. The spleen of man, however, has a fibro-elastic capsule without any smooth muscle fibers and the trabeculae possess only an occasional muscle band. From its anatomical description, the human spleen is potentially a much less contractile organ than that in the dog.

Recently Wright, Doan and co-workers presented an extensive and detailed study on the effect of epinephrine injected into the splenic artery in man, and demonstrated a marked rise in leukocytes in the splenic vein in patients with idiopathic thrombocytopenic purpura and hypersplenism. On the basis of these findings, the authors concluded that the peripheral blood leukocytosis following epinephrine is due to release of leukocytes from the spleen. This conclusion is used as definitive support for the epinephrine test, in which the functional capacity of the spleen is measured by the leukocyte levels in the blood following intravenous or subcutaneous injection of epinephrine, and for the physiologic concept of the spleen as a major reservoir of leukocytes as well as in the pathologic concept of the syndrome of hypersplenism.

Critical consideration of the data of Wright et al however, indicates that the data fail to support the conclusions. Samples of splenic vein blood obtained 2 to 5 minutes following the injection of 0.3 to 1.0 mg. of epinephrine into the splenic artery, showed increases in leukocyte number which, although marked in some instances, could not account for the leukocytosis usually observed in the peripheral blood. In 18 cases in which control splenic artery and vein blood samples were available, the average increase in leukocyte number in the splenic vein blood following epinephrine was about four times that originally observed in the splenic artery. To double the peripheral leukocyte count in an average individual with a blood volume of 6,000 cc. would require 1,500 cc. of blood with the maximum number of leukocytes continuously entering the peripheral circulation within 2 to 5 minutes. Furthermore, these calculations ignore any loss of cells in transit through other capillary beds and assume the prompt and complete release of cells from the spleen although the peak leukocytosis occurred momentarily and at 15 to 20 minutes in the 2 cases in which comparative data with peripheral blood was available. In 7 cases of the primary hypersplenic syndromes in which granulocyte counts from the splenic vein were available before and after intra-arterial splenic epinephrine was administered, the counts averaged 9,100 and 41,000 respectively. This increase of granulocytes by a multiple of 4.5 is again insufficient to account for the increases in peripheral blood granulocytes solely from the spleen.

Similarly, in the 9 patients with essential thrombocytopenic purpura listed under the primary hypersplenic syndrome the average pre-epinephrine platelet count in the splenic vein samples was 59,000 and the average post-epinephrine platelet count was 122,000. This increase alone from the spleen would not be reflected significantly in the peripheral blood. It must be emphasized that the
patients of Wright et al. had primary hematologic dyscrasias and that excessive concentrations of leukocytes or platelets may have been released but no data to support such a contention were presented. The average volume of the spleens in the patients reported could reasonably be expected not to exceed 1,000 cc., or less than the volume of the blood necessary to be added to the circulating volume to cause the leukocytosis in the peripheral blood. It is, therefore, impossible to account for the leukocytosis exclusively on release of cells from the spleen, even if the total volume of the spleen was all blood. These data make it obvious that the leukocytosis following epinephrine must include other major sources of cells in addition to the spleen. Furthermore, the leukocyte reaction can hardly be a reliable test for the functional activity of the spleen under all circumstances since it is produced in its absence.

The human spleen will decrease in size within 2 to 5 minutes following the administration of epinephrine directly into the splenic artery. The administration of 0.2 to 0.4 mg. of epinephrine directly into the splenic artery is followed within 30 seconds by marked and almost complete arterial constriction a few centimeters distal to the point of administration. This was further confirmed by the absence of any arteriographic pattern distal to the constricted point for 1 to 2 minutes after injection and the inability to withdraw blood samples from the catheter in the splenic artery during this period of time. The splenic artery contraction is prolonged and apparently so effective in shutting off the arterial blood flow that Wright et al. also reported difficulty in aspiration of even small amounts of blood.

A significant arterial leukocytosis following the parenteral administration of epinephrine has been observed in man presumably from a leukocyte reservoir in the pulmonary circulation. Since a leukocytosis following epinephrine occurs in the absence of the spleen, it became desirable to define the specific roles of both the lung and spleen following epinephrine administration. In an effort to obtain a baseline for future investigations in patients with various hematologic dyscrasias and with the nonavailability of patients with hypersplenism, patients with various neoplastic diseases were studied.

**Subjects and Method**

Surgical procedures involving a laparotomy, general anesthesia, manipulation of the spleen or splenic pedicle may influence the leukocyte and platelet count unduly and, consequently, the conventional operative approach was avoided. The technics of simultaneous intra-arterial and intravenous catheterization were employed to study this problem in the intact patient.

Seven patients with advanced neoplastic diseases were studied in the following manner. A No. 9 or 10F arterial catheter was passed from the left brachial artery into the descending aorta and then manipulated into the splenic artery by the method previously described. Five cubic centimeters of thorotrast or Neo-iopax were injected rapidly through the catheter and a film exposed at the proper time demonstrated the characteristic splenic arteriographic pattern. A similar catheter was simultaneously introduced via the right basilic or cephalic vein and manipulated into either the hepatic vein or the pulmonary artery.

With both catheters in place, an indwelling needle was placed percutaneously into the femoral or brachial artery from which adequate samples could be obtained. Epinephrine* as adrenalin chloride 1:1000 solution diluted directly from the ampoule in 10 cc. of isotonic saline.
### Table 1—Hematologic Data on 7 Patients Receiving Intra-arterial Epinephrine

<table>
<thead>
<tr>
<th>Name</th>
<th>Age</th>
<th>Sex</th>
<th>Diagnosis</th>
<th>Dose Epinephrine mg.</th>
<th>Sample Sites</th>
<th>Control WBC</th>
<th>Peak WBC</th>
<th>Time in seconds</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>Car</td>
<td>37</td>
<td>M</td>
<td>Malignant melanoma</td>
<td>0.3</td>
<td>Pulmonary Conus</td>
<td>1,400</td>
<td>2,800</td>
<td>270 300</td>
<td>Artery leukocyte count rose in 90 sec. and venous blood did not show rise until 270 sec. Granulocytes were primarily involved.</td>
</tr>
<tr>
<td>You</td>
<td>49</td>
<td>M</td>
<td>Malignant melanoma</td>
<td>0.3</td>
<td>Pulmonary Conus</td>
<td>5,700</td>
<td>6,700</td>
<td>270 360</td>
<td>Arterial leukocyte rise at 120 sec. mainly in granulocytes with slight rise in venous samples at 240 sec.</td>
</tr>
<tr>
<td>Cas</td>
<td>54</td>
<td>M</td>
<td>Monocytic leukemia</td>
<td>0.2</td>
<td>Pulmonary Conus</td>
<td>3,000</td>
<td>5,000</td>
<td>120 210</td>
<td>Arterial rise preceded the venous rise. Both attained the same height.</td>
</tr>
<tr>
<td>Ley</td>
<td>46</td>
<td>M</td>
<td>Carcinoma of esophagus</td>
<td>0.2</td>
<td>Pulmonary Conus</td>
<td>800</td>
<td>1,200</td>
<td>185 300</td>
<td>Patient had marked leukopenia during study with counts between 800 and 1,200 with slight changes.</td>
</tr>
<tr>
<td>Mur</td>
<td>34</td>
<td>F</td>
<td>Malignant melanoma</td>
<td>0.2</td>
<td>Hepatic vein</td>
<td>4,600</td>
<td>7,700</td>
<td>120 180</td>
<td>No rise in hepatic vein samples until 120 sec. due exclusively to granulocytes. Arterial rise in granulocytes of from 5,100 to 6,000 in first 30 sec.</td>
</tr>
<tr>
<td>Goo</td>
<td>27</td>
<td>M</td>
<td>Anaplastic carcinoma</td>
<td>0.2</td>
<td>Hepatic vein</td>
<td>900</td>
<td>3,000</td>
<td>60 270</td>
<td>Arterial rise in granulocytes preceded the venous rise. Both attained the same height.</td>
</tr>
<tr>
<td>Man</td>
<td>40</td>
<td>F</td>
<td>Carcinoma of lung</td>
<td>0.2</td>
<td>Pulmonary Conus</td>
<td>8,400</td>
<td>12,100</td>
<td>90 180</td>
<td>Arterial counts were higher than the vein counts throughout the whole study.</td>
</tr>
</tbody>
</table>

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in doses of 0.2 to 0.3 mg. was administered through the arterial catheter into the splenic artery at a uniform rate over a 30 second period with precautions to avoid even the slightest reflux into the aorta. The administration of 10 cc. of contrast medium over a period of 3 to 5 seconds under identical conditions failed to exhibit any reflux into the aorta. In 1 patient, the epinephrine was administered in the aorta at a point distal to the splenic artery. Arterial and venous blood samples were obtained simultaneously at 30 second intervals during and following the epinephrine administration for 5 minutes and then every minute for an additional 5 to 10 minutes.

Erythrocyte, leukocyte and platelet counts were determined with National Bureau of Standards certified hemoctometers and Treiner automatic filling pipets. Heparin was employed as the anticoagulant and all pipets were filled directly from the original samples in the collecting tubes and were shaken uniformly for 10 minutes in an automatic rotator. At least 600 cells on all squares on both chambers were counted and duplicating chambers were determined by the direct method employing Tocantin’s solution. The over-all standard error for counting leukocytes was estimated at less than ±8 per cent at 70 per cent confidence limits for counts between 5,000 to 10,000 per cu. mm.

RESULTS

Epinephrine, 0.2 or 0.3 mg., was introduced into the splenic artery via an intra-arterial catheter in 6 patients with far advanced neoplastic diseases (table 1). In 2 patients, samples of hepatic vein and peripheral arterial blood were taken simultaneously during and following the administration of epinephrine (table
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1). In the 4 other patients the venous samples were obtained from the pulmonary conus. In 1 additional patient, the epinephrine was administered intra-arterially (aorta) at a point distal to the celiac artery. Within 30 to 60 seconds after starting the injection of epinephrine an initial rise in leukocyte count occurred in the peripheral arterial blood which was further accentuated at 2 to 3 minutes. The samples from the hepatic vein or pulmonary conus showed an

Fig. 2A—Mur, female, age 34. Rise in leukocyte number in arterial blood from 4,000 to 6,000 in first 30 seconds. Venous samples did not show any significant change until 120 seconds and then exceeded the number of leukocytes in the arterial blood for the next 100 seconds. The changes were exclusively in the granulocytic series.

Fig. 2B—Catheter in splenic artery with arteriogram illustrating splenic pattern. Figure to the right shows catheters in hepatic vein and splenic artery.
initial rise in leukocyte count at 120 to 180 seconds following the start of the epinephrine injection into the splenic artery (fig. 1). In 3 of the 6 patients in whom epinephrine was administered into the splenic artery, the changes in the arterial blood preceded those in the venous blood by 90 to 180 seconds.

The rise in leukocyte number persisted for approximately 3 minutes and then gradually subsided. The leukocyte count in patient Mur in samples obtained directly from the hepatic vein exceeded that in the peripheral arterial blood by approximately 1,000 cells/cu. mm. for a short period following the initial rise after which the arterial blood leukocyte number exceeded that in the hepatic vein (fig. 2). When the venous samples were derived from the pulmonary conus, however, the increase in leukocyte number was smaller than obtained in samples from the hepatic vein and never significantly exceeded the number in the arterial blood in the first 5 minutes (fig. 3). The increase in leukocytes in the arterial blood

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**Fig. 3**—You, male, age 49. Prompt arterial leukocyte rise exceeding the venous count which initially was granulocytic, joined one minute later by an increase in lymphocytes. The significant venous rise does not occur until 90 seconds after the initial arterial rise.
occurred predominantly in the granulocytes while the pulmonary conus blood revealed its increase in both granulocytes and agranulocytes.

Fig. 4A  Catheter tip in left pulmonary artery introduced from the saphenous vein. The arterial catheter can be seen in the aorta which passes from the left brachial into the splenic artery.

B  Cas. male, age 54. Splenic arteriogram with 12 cc. thorotrast.

The spleen size was observed to decrease measurably within 40 seconds after the initiation of 0.2 mg. epinephrine administration into the splenic artery (figs. 4A, B, C). The small vessel pattern was absent and the larger arteries exhibited
C—Splenic arteriogram with 12 cc. thorotrac 40 seconds after 0.2 mg. epinephrine into the splenic artery with identical technic used in the study of circulating leukocytes.

D—Cas, male, age 54. Increase in granulocytes in arterial blood preceding those in the venous blood following intra-arterial splenic administration of epinephrine.

marked diminution in size. The leukocyte counts in the pulmonary artery samples in this patient (Cas) following 0.2 mg. epinephrine showed no rise until 120 seconds consisting entirely of granulocytes and did not exceed the rise in the arterial blood (fig. 4D).
The injection of epinephrine into the aorta distal to the celiac artery was followed by prompt increases in leukocytes in the arterial blood as occurred with intravenous epinephrine administration and did not differ essentially from the 6 patients in whom the epinephrine was given into the splenic artery. There was a consistent downward trend in the platelet number in the venous samples while simultaneously the arterial blood platelet number was maintained or increased (fig. 1). The systemic reaction of the patients to the intrasplenic epinephrine infusion was delayed and less severe when compared with the usual response following intravenous injection of identical amounts of epinephrine.

The characteristic subjective and objective sensations of epinephrine administration were usually not manifest until approximately 60 seconds after the start of the injection.

**DISCUSSION**

The pulmonary circulation has been shown to act as an immediate source of leukocytes and platelets following intravenous epinephrine administration.\(^2\) The intra-arterial splenic administration of epinephrine was followed by an immediate rise in leukocytes in the peripheral arterial blood which persisted for approximately 5 minutes. The leukocytosis from the spleen and liver did not initially appear until at least 120 seconds after the epinephrine injection trailing the increase in leukocytes in the arterial blood by at least two or more circulation times. It should be emphasized that the doses of epinephrine injected directly into the splenic artery were far in excess of what the spleen would receive via the subcutaneous route. The peripheral blood leukocytosis following the intrasplenic epinephrine administration, therefore, probably comes from at least two sources; initially from the pulmonary circulation, accentuated 2 to 3 minutes later by an increased number of leukocytes from the spleen and other sites. Consequently, the leukocytosis following the parenteral administration of epinephrine per se cannot be used as a reliable measure of splenic contractility or sequestration. This does not necessarily exclude the relationship of the leukocyte response to various hematologic pathoses but rather emphasizes the possible role of the lung in these conditions.

Since the venous drainage of the spleen is via the portal circulation through the liver, the hepatic vein contains those blood elements from the entire portal drainage including the spleen and gastro-intestinal tract. It must be emphasized that increased numbers of leukocytes leaving the splenic vein would be diluted by the entire circulating blood volume which would require extremely large and protracted increases in leukocyte number in the splenic vein blood to be reflected significantly in the peripheral blood provided they all successfully pass through the hepatic and then the pulmonary circulation. Consequently, the hepatic vein counts should have been higher than those sampled from the pulmonary artery and such were the findings. Similarly, the magnitude of the changes in the venous blood in the two instances in which the hepatic vein blood was sampled exceeded those in the arterial blood. In all but one of the remaining instances the converse was true.

Barcroft and Poole\(^2\) were unable to account for the changes in blood following contraction of the spleen entirely on the decrease in size of the spleen and
suggested other sources of cells. The apparent constriction of the splenic artery following direct epinephrine administration and the comparative absence of smooth muscle in the spleen suggests that the decrease in size of the human spleen is not primarily due to an active contraction of the splenic capsule, trabeculae or any other part of the splenic parenchyma except the vasculature.

It is postulated that under these circumstances epinephrine given intra-arterially into the spleen causes constriction of the splenic artery temporarily reducing the flow of arterial blood into the spleen for 1 to 2 minutes. The effect of such an infusion would be manifested throughout the spleen involving the responsive arteries and arterioles with progressively less direct influence if any on the capillary bed or venous circulation. Consequently, the splenic vascular bed may empty with shrinkage of the spleen by exsanguination rather than by active contraction of the splenic substance. With the release of the arterial spasm, the blood flow through the spleen is re instituted and leukocytes from within the splenic vessels are flushed into the vein to accentuate further the immediate leukocytosis from the lungs. This mechanism of leukocytosis may not pertain to those patients with pathologic conditions differing from those studied here.

It should be emphasized that none of the patients studied here had any primary disease of the spleen insofar as could be determined.

CONCLUSIONS

1. The injection of 0.2 to 0.3 mg. of epinephrine directly into the splenic artery by means of an intra-arterial catheter in 6 patients with various neoplastic diseases was followed by a leukocytosis of both granulocytes and agranulocytes in blood from the hepatic vein or pulmonary artery which first appeared about 120 to 270 seconds after injection and persisted for approximately 3 minutes. There was no increase in platelet number in the venous blood samples.

2. In 1 additional patient, epinephrine administered arterially at a point distal to the splenic artery resulted in a similar leukocytosis.

3. The leukocytosis in the peripheral arterial blood was primarily granulocytic and appeared within 30 to 60 seconds after start of the injection.

4. The increase in platelets in the arterial blood exceeded the number of platelets in the venous blood in 4 of the 6 patients.

5. It is concluded that the leukocytosis in these patients following the administration of epinephrine into the splenic artery is supplied from at least two sources; the immediate increase of leukocytes from the pulmonary circulation with accentuation by the release of leukocytes from the spleen and possibly the other portal organs 2 to 5 minutes later.

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