Megakaryocytes in the Pulmonary Circulation

By T. M. Scheinin and A. Koivuniemi

The frequent occurrence of circulating megakaryocytes has only recently been proved. Histologic studies have repeatedly shown large numbers of these cells in the pulmonary capillaries. Next to bone marrow, the lungs appear to be the most frequent site of the megakaryocytes. The role of the pulmonary megakaryocytes as a source of blood platelets has been disputed. Whereas Howell and Donahue and Sharnoff and Kim considered these cells to be of importance in the production of platelets, this concept has not been shared by others.

During the past few years the megakaryocytes have been the focus of considerable interest because of their importance in the differential diagnosis of circulating cancer cells. The various methods aimed at effective isolation of cancer cells in the blood are based on the elimination of many of the benign blood cells. The remaining cellular smear contains, however, a number of megakaryocytes which can be recognized without too much difficulty. The methods for isolation of cancer cells thus offer possibilities for studies of circulating megakaryocytes.

This work was undertaken in order to study the incidence and characteristics of megakaryocytes in circulating blood of the lungs in patients with malignant and nonmalignant pulmonary disease.

Material and Method

The essential part of this series consisted of 42 patients with histologically proved lung cancer and of 21 patients with nonmalignant inflammatory disease of the lungs. In these cases blood samples were taken (1) from the pulmonary artery upon opening of the chest wall, (2) from the pulmonary vein before manipulation of tumor or diseased lung, and (3) from the pulmonary vein during manipulation of tumor or lung. In addition, the occurrence of megakaryocytes in peripheral antecubital venous blood was studied in 72 cases of malignant tumor in various organs and in 33 cases with nonmalignant disease (22 cases of inflammatory disease or severe anemia, and 11 cases of other nonmalignant disease). In eight cases samples were taken from peripheral arterial blood.

The blood samples varied from 4 to 10 ml. The enzymatic streptolysin O hemolysis method, a modification of the technic reported by Malngren and his associates, was employed for the isolation of the megakaryocytes. The cellular smears were stained by the Papanicolaou technic.

Control examinations were made in order to test the effect of the streptolysin technic on megakaryocytes from the bone marrow. The cells appeared to be well preserved and without loss of cytoplasm or nuclear characteristics.
MEGAKARYOCYTES IN THE PULMONARY CIRCULATION

Table 1.—Megakaryocytes in the Pulmonary Circulation and Effect of Surgical Manipulation of Lung

<table>
<thead>
<tr>
<th>Time and Site of Blood Sampling</th>
<th>Circulating Megakaryocytes</th>
<th>Megakaryocytes with Preserved Cytoplasm</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No. Cases</td>
<td>%</td>
</tr>
<tr>
<td>Pulmonary artery at thoracotomy</td>
<td>48</td>
<td>76</td>
</tr>
<tr>
<td>Pulmonary vein at thoracotomy</td>
<td>28</td>
<td>44</td>
</tr>
<tr>
<td>Pulmonary vein during manipulation</td>
<td>41</td>
<td>65</td>
</tr>
</tbody>
</table>

RESULTS

In the total series of 168 patients, megakaryocytes were found in 129 cases, an incidence of 77 per cent. The blood samples contained, on an average, 1.2 megakaryocytes per ml. These cells were definitely more frequent in pulmonary arterial blood than in pulmonary venous blood (table 1). In this respect there was no difference between the lung cancer group and the non-malignant group. Most of the circulating megakaryocytes appeared thus to be arrested in the pulmonary vascular bed and only a relatively small part of them reached into the venous blood. The pulmonary venous samples contained generally smaller megakaryocytes with a greater share of nuclear fragments than the pulmonary arterial samples. Manipulation of tumor-bearing or inflamed lung tissue increased the number of these cells in pulmonary venous blood.

The circulating megakaryocytes were generally without cytoplasm or with only a narrow rim of it. Only 9 per cent of the total number of cells had an abundant cytoplasm (figs. 1 and 2). Nearly all of these large intact cells were seen in samples from pulmonary arterial blood (table 1). It seemed that the cells lost most or all of their cytoplasm while passing from pulmonary arterial to pulmonary venous blood.

The antecubital venous blood samples showed greater numbers of megakaryocytes (75 per cent positive findings) and of cells with preserved cytoplasm (7 per cent of the cases) than the pulmonary venous blood samples.

As shown in table 2, the megakaryocytes were more frequent in advanced malignant disease than in the early cases. They were more common in inflammatory disease and severe anemia than in other non-malignant disease.

DISCUSSION

In this series megakaryocytes and their nuclear fragments were frequently found in blood samples from the pulmonary vessels. They were most common in pulmonary arterial blood, whereas pulmonary venous blood contained markedly smaller numbers of these cells. This difference was in our opinion too great to be accounted for by possible changes in the plasma content of the blood during passage through the lungs. Operative manipulation of the lung seemed to increase the amount of pulmonary venous megakaryocytes.

The findings consisted mostly of bare megakaryocytic nuclei or their frag-
ments. The pulmonary venous samples contained a particularly large fraction of smallish, naked megakaryocyte nuclei. Whenever megakaryocytes had an intact abundant cytoplasm they almost invariably were found in the pulmonary arterial blood samples.

Antecubital venous blood contained megakaryocytes in a greater percentage of cases than the pulmonary venous blood. The incidence of megakaryocytes in the peripheral circulation in our series agrees fairly well with some recent observations. In blood samples from our cancer group, megakaryocytes were more frequent in advanced disease than in early cases. In nonmalignant cases they were more numerous in inflammatory disease or blood dyscrasia than in other benign conditions. This finding corresponds to observations that circulating megakaryocytes are more frequent in diseases associated with increased bone marrow activity.

Histologic studies have demonstrated the frequent occurrence of megakaryocytes in the lungs, where they generally are confined to the capillaries of the alveolar septa. The prevailing opinion, mainly based on indirect evidence, seems to be that the megakaryocytes of the lungs are migrating cells from the bone marrow which have been trapped in the pulmonary capillary bed. The present study gives more direct evidence of the filtering function of the lungs in humans. On the bases of this series and previous histologic evidence, it appears that the circulating megakaryocytes arrive in small numbers to the lungs with the pulmonary arterial blood. The cells are trapped and deformed in the lung capillaries. A small fraction of the cells have an abundant cytoplasm when they reach the lungs, whereas only very few of the
Megakaryocytes in pulmonary venous blood have retained their cytoplasm. It is possible that the pulmonary megakaryocytes contribute to the formation of platelets, but the significance of this source of blood platelets is open to discussion.11,15

Relatively few megakaryocytes were found in pulmonary venous blood and many of them were in fact nuclear fragments. This might be the result of filtration, degeneration and subsequent fragmentation of the arrested cells in the lung capillaries.

Summary

The streptolysin 0 hemolysis method for isolation of cancer cells in the blood was employed for direct observations of the incidence and some characteristics of circulating megakaryocytes.

In a series of 168 patients, circulating megakaryocytes were found in 77 per cent of the blood samples. Each sample contained an average of 1.2 megakaryocytes per ml. of blood. The megakaryocytes were most frequent in pulmonary arterial blood and a number of the cells had an apparently intact

Table 2.—Incidence of Circulating Megakaryocytes According to Disease

<table>
<thead>
<tr>
<th>Disease</th>
<th>No. of Megakaryocytes per Sample</th>
<th>No. of Megakaryocytes with Preserved Cytoplasm per Sample</th>
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</thead>
<tbody>
<tr>
<td>Total cancer series</td>
<td>6.2</td>
<td>0.6</td>
</tr>
<tr>
<td>Advanced cases</td>
<td>9.2</td>
<td>0.8</td>
</tr>
<tr>
<td>Early cases</td>
<td>3.3</td>
<td>0.3</td>
</tr>
<tr>
<td>Inflammatory disease and severe anemia</td>
<td>4.5</td>
<td>0.3</td>
</tr>
<tr>
<td>Other benign conditions</td>
<td>1.0</td>
<td>—</td>
</tr>
</tbody>
</table>
abundant cytoplasm. Pulmonary venous blood contained megakaryocytes much less frequently. These were almost always without cytoplasm or with only a narrow rim of it and as a rule small naked nuclei or their fragments were found. Manipulation of lung tissue resulted in an increased amount of megakaryocytes in the pulmonary venous blood.

The megakaryocytes in pulmonary and systemic circulation were more numerous in advanced malignant disease than in early cases, and more common in inflammatory disease or severe anemia than in other nonmalignant disease.

SUMMARIO IN INTERLINGUA

Le metodo del hemolyse per streptolysina O pro le isolation de cellulas de cancere in le sanguine eseva emplete pro le directe observation del incidentia e de certe caracteristicas de megacaryocytos in le circulation.

In un serie de 168 patientes, circulante megacaryocytos eseva trovate in 77 pro cento del specimens de sanguine. Omne specimen contineva como valor medie 1,2 megacaryocytos per ml de sanguine. Le megacaryocytos eseva le plus frequente in sanguine de arteria pulmonar, e un numero del cellulas habeva un apparentemente intacte e abundante cytoplasma. Sanguines de vena pulmonar contineva megacaryocytos multo minus frequentemente. Isto eseva quasi semper disproviste de cytoplasma o possedente solmente un fin margine di illo. Como regula general, micre nude nucleos o fragmentos de illos eseva trovate. Manipulation del tissu pulmonar resultava in un augmentate frequentia di megacaryocytos in le sanguine de vena pulmonar.

Le megacaryocytos in le circulation pulmonar e general eseva plus numerose in avantiate casos de morbo maligne que in casos precoce. Illos eseva plus commun in morbo inflammatori o in anemia sever que in altere morbos de typo non maligne.

REFERENCES

11. Sharnoff, J. G., and Kim, E. S.: Evalua-
13. Sharnoff, J. C., and Kim, E. S.: Pulmonary megakaryocyte studies in rab-
14. Jordan, H. E.: The origin and significance of the megakaryocytes of
15. Fidlar, E., and Waters, E. T.: The origin of platelets, their behavior in
nation of Cancer: Prevention and Therapy. New York, Appleton-Cen-
36:1, 1922.
21. Whitby, L.: The significance of megakaryocytes in the peripheral circula-

T. M. Scheinin, M.D., Department of Thoracic Surgery, Central
University Hospital, Helsinki, Finland.

A. P. Koivuniemi, M.D., Section II of the Department of Path-
ology, University of Helsinki, Helsinki, Finland.
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T. M. SCHEININ and A. P. KOIVUNIEMI