

Clinical correlates of splenic histopathology and splenic karyotype in myelofibrosis with myeloid metaplasia

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Splenic extramedullary hematopoiesis is an integral component of myelofibrosis with myeloid metaplasia (MMM) and may be classified into 3 distinct histologic patterns of infiltration by myeloid precursors: diffuse, nodular, and a predominance of immature granulocytes. These 3 histologic patterns occurred in 121 (56.8%), 75 (35.2%), and 17 (8%), respectively, of 213 patients with MMM who underwent splenectomy at a single

institution. In general, karyotypic findings in splenic tissue (n = 92) were similar to those seen in the bone marrow. The histologic pattern of immature granulocyte predominance, the presence of microscopic splenic infarcts (26 patients), or the detection of an abnormal splenic karyotype (52 patients) was significantly associated with decreased postsplenectomy survival. These adverse features were also associated with charac-

teristics of advanced disease. These observations support the bone marrow origin of the myeloid progenitor pool in the spleen of patients with MMM and suggest a prognostic value for splenic histopathology and karyotype. (Blood. 2001;97:3665-3667)

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Introduction

Specific extramedullary hematopoiesis (EMH) in myelofibrosis with myeloid metaplasia (MMM) was originally believed to arise from reactivation of fetal hematopoietic elements.¹ Current evidence suggests that splenic EMH in MMM results from sequestration, accumulation, and proliferation of circulating myeloid progenitors in splenic cords.² Immunohistochemical analysis of splenic tissue has revealed that EMH in MMM is primarily granulocytic³ as compared with the fetal spleen, which is mainly a site for erythroid differentiation.⁴ The bone marrow origin of EMH precursors in MMM has further been suggested by immunohistologic and morphometric studies of megakaryocytes⁵ and the in vitro demonstration of committed, but not pluripotent, myeloid progenitors in splenic tissue.⁶ Splenectomy may be necessary to palliate symptoms and improve the quality of life in patients with MMM.⁷ It was recently reported that splenic pathology in MMM may undergo a prognostically relevant progression from an erythroid to a panmyeloid composition.⁸ Accordingly, and to provide complementary information in a recently described series of patients with MMM who underwent splenectomy,⁷ we investigated the prognostic value of splenic histopathology and karyotype.

the presence or absence of microscopic splenic infarctions was noted (Figure 1), and the results of karyotypic studies in the splenic tissue and bone marrow were recorded.

Correlations among clinical, histologic, and cytogenetic parameters were studied by nonparametric statistical techniques. The relations between categorical variables were studied with the Fisher exact test. When a continuous variable was divided into 2 categories, the Wilcoxon rank sum test was used to compare the medians of the continuous variable between the 2 categories. When a continuous variable was divided into 3 or more categories, medians of the continuous variable in each of the 3 or more categories were compared by means of a Kruskal-Wallis test. Kaplan-Meier⁹ methodology was used to estimate the distributions of survival from diagnosis and survival from splenectomy. The log-rank test was used to assess whether survival from diagnosis and survival from splenectomy differed between various categories. Multivariate analysis was performed using logistic regression.

Study design

Archived splenic tissue obtained from splenectomized patients with MMM (n = 213) was stained with hematoxylin and eosin and examined under light microscopy by one of the authors (C.-Y.L.), who was blinded to the clinical characteristics and the outcomes of the study patients. All cases were histologically categorized according to the pattern of myeloid precursor infiltration in the spleen. The 3 categories recognized were diffuse (diffuse pattern of EMH with trilineage myeloid involvement), nodular (macronodular proliferation of EMH), and a predominance of immature granulocytes (immature granulocyte predominance) (Figure 1). In addition,

Results and discussion

One hundred twenty-one (56.8%) of the 213 splenectomized patients with MMM had a diffuse infiltrative pattern of EMH that was composed of granulocytic, erythroid, and megakaryocytic precursors (diffuse). In another 75 patients (35.2%), this trilineage infiltration of precursors formed a macronodular pattern (nodular). The histologic pattern in the remaining 17 patients (8.0%) consisted almost exclusively of immature granulocyte precursors (immature granulocyte predominance). In none of the patients was the EMH composed strictly of erythroid elements. Results of our histologic review of MMM splenic tissue are consistent with those of previous reports.^{3,10} Patients with immature granulocyte predominance had unfavorable prognostic scores¹¹ ($P = .04$) and a higher incidence of cytopenias (erythrocyte transfusion dependence,

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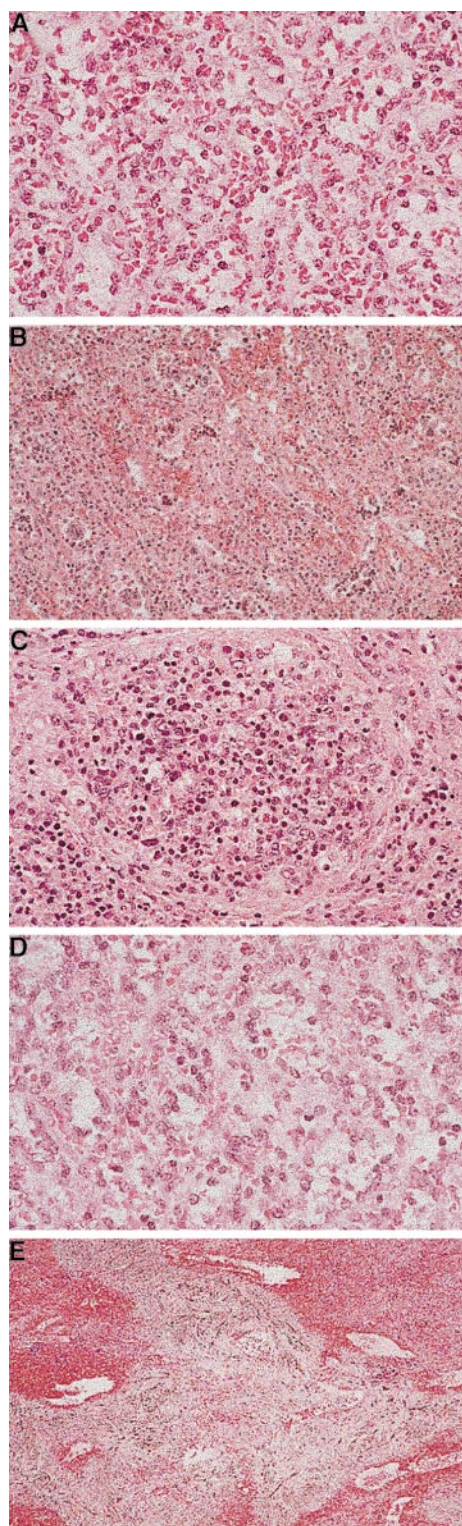


Figure 1. Splenic histologic findings in patients with myelofibrosis with myeloid metaplasia. (A) Normal spleen ($\times 128$). (B) Diffuse splenic EMH ($\times 128$). (C) Nodular splenic EMH ($\times 80$). (D) Immature granulocytic predominant EMH ($\times 128$). (E) Splenic microinfarction ($\times 51$).

$P < .01$; and platelet count less than $50 \times 10^9/L$, $P < .01$ compared with those with more balanced trilineage EMH (nodular or diffuse) (Table 1). In addition, independent of blastic transformation, the particular histologic pattern was associated with decreased overall survival (Figure 2A). The respective median survival times

from diagnosis were 49.4, 64.5, and 90.8 months for immature granulocyte predominance, diffuse, and nodular EMH histology. Although direct comparison of the 2 most common histologic patterns revealed no survival difference, patients with diffuse EMH had significantly worse prognostic scores¹¹ and a higher incidence of cytopenias (Table 1). Collectively, these observations suggest that splenic EMH in MMM may initially follow a nodular pattern and then undergo a prognostically relevant histologic transformation into a diffuse pattern first and granulocyte predominance second.

Microscopic splenic infarcts were observed in 26 patients and were not associated with a particular histologic pattern or with the occurrence of postsplenectomy thrombocytosis or vascular events. In contrast, their presence was significantly associated with an adverse prognostic score,¹¹ thrombocytopenia, and the presence of circulating blasts. In addition, patients with splenic infarcts were

Table 1. Results of statistical analysis among splenic histologic subgroups in 213 splenectomized patients with myelofibrosis with myeloid metaplasia

Parameter	Pathology category (n = 213)	Diffuse vs nodular (n = 196)	Splenic infarct (n = 26)
General parameters			
Number of patients	D = 121 (56.8%) N = 75 (35.2%) PPIG = 17 (8.0%)	D = 121 (61.7%) N = 75 (38.3%)	26 (12.2%)
Survival			
From diagnosis	.04	.17	.01
From splenectomy	.29	.82	.02
Cause of death	.39	.14	.85
Dupriez score*			
At diagnosis	.09	.02†	< .01
At splenectomy	.04	.01	.20
Time to splenectomy (from diagnosis)			
	.82	.16	.18
Parameters at the time of splenectomy			
Age	.99	.53	.38
Laboratory values			
Hemoglobin < 9 g/dL	.20	.10	.09
RBC transfusion dependent (yes/no)	< .01	.02	.09
Leukocyte count	.27	.49	.16
Platelet count < $50 \times 10^9/L$	< .01	< .01†	< .01†
Circulating blasts	.44	.44	.04
Spleen mass	.70	.10	.63
Splenic infarct (yes/no)	.90	.56	—
Perioperative complications			
Bleeding	.87	1.0	.02
Thrombosis	1.0	1.0	.23
Long-term complications			
Leukemia	.65	.75	.08
Thrombocytosis	.27	1.0	.62
Hepatomegaly	.70	.43	.77

All results (except those for distribution) reflect a P value from univariate analysis. Column 1, D vs N vs PPIG; column 2, D vs N only.

D indicates diffuse extramedullary hematopoiesis pattern; N, nodular extramedullary hematopoiesis pattern; PPIG, predominant presence of immature granulocytes in splenic extramedullary hematopoiesis; RBC, red blood cell.

*Dupriez prognostic score for myelofibrosis with myeloid metaplasia.¹¹

†Significant in multivariate analysis using logistic regression.

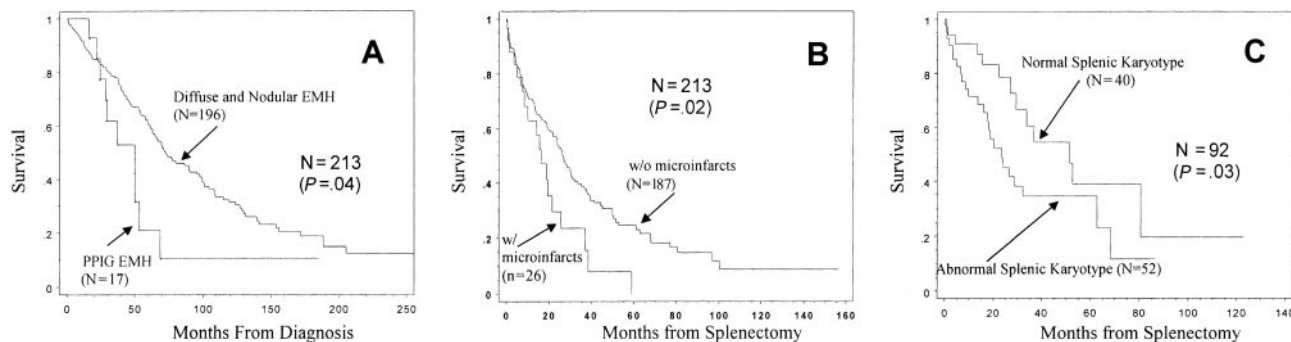


Figure 2. Histologic prognostic factors from splenic tissue in 213 patients with myelofibrosis with myeloid metaplasia. (A) Survival from diagnosis according to splenic EMH. (B) Survival from splenectomy according to the presence of microinfarctions. (C) Survival from splenectomy according to splenic karyotype. PPIG indicates predominance of immature granulocytes.

more likely to have their disease transform into acute leukemia ($P = .08$) and to have worse overall and postsplenectomy survival (Figure 2). Splenic cytogenetic studies were performed in 92 patients, and 52 (56.5%) had an abnormal karyotype (29 single and 23 multiple karyotypic lesions). Specific abnormalities included $20q-$ ($n = 15$), $13q-$ ($n = 11$), $+9$ ($n = 5$), abnormalities of chromosome 5 or 7 ($n = 5$), $12p-$ ($n = 4$), abnormal chromosome 1 ($n = 4$), isochromosome 17q ($n = 3$), and $+8$ ($n = 2$). Of the 92 patients who had splenic karyotype analysis, 68 had information on bone marrow karyotype that was performed either before ($n = 60$) or after ($n = 8$) splenectomy. The karyotypic findings in the 2 tissues were concordant in more than 85% of cases. Among 9 of the 10 patients who had karyotype discordance between spleen and bone marrow, the splenic karyotype showed the same clone as in the bone marrow but with additional chromosomal lesions. Only one patient had an abnormal karyotype that was found in the bone marrow but not in the spleen. The presence of an abnormal splenic

karyotype was associated with decreased postsplenectomy survival ($P = .03$) (Figure 2), but not with other clinicopathologic variables.

The excellent concordance between bone marrow and splenic cytogenetic clones, as well as our histologic observations, strengthens the hypothesis¹² that splenic EMH in MMM arises from filtration of the clonally involved circulating progenitor cells. Splenic EMH arising in other conditions, such as myelophthisis from metastatic cancer or marrow stimulation by granulocyte colony-stimulating factor,¹³ has also been shown to arise from filtration of circulating progenitors.^{14,15} The peripheral blood progenitor pool in MMM is markedly elevated,¹⁶ and its preferential localization and proliferation in the spleen and liver suggest that these organs provide an environment conducive to progenitor growth and differentiation. Our observation concerning the detrimental prognostic significance of immature granulocyte predominance of splenic EMH may therefore reflect a changing circulating progenitor pool in advanced MMM.

References

- Dameshek W. Some speculations on the myeloproliferative syndromes [editorial]. *Blood*. 1951;6:372-375.
- Zhang B, Lewis SM. The splenomegaly of myeloproliferative and lymphoproliferative disorders: splenic cellularity and vascularity. *Eur J Haematol*. 1989;43:63-66.
- Wilkins BS, Green A, Wild AE, Jones DB. Extramedullary haematopoiesis in fetal and adult human spleen: a quantitative immunohistological study. *Histopathology*. 1994;24:241-247.
- Calhoun DA, Li Y, Braylan RC, Christensen RD. Assessment of the contribution of the spleen to granulocytopenia and erythropoiesis of the mid-gestation human fetus. *Early Hum Dev*. 1996;46:217-227.
- Thiele J, Klein H, Falk S, Bertsch HP, Fischer R, Stutte HJ. Splenic megakaryocytopenia in primary (idiopathic) osteomyelofibrosis: an immunohistological and morphometric study with comparison of corresponding bone marrow features. *Acta Haematol*. 1992;87:176-180.
- Douay L, Laporte JP, Lefrancois G, et al. Blood and spleen haematopoiesis in patients with myelofibrosis. *Leuk Res*. 1987;11:725-730.
- Tefferi A, Mesa RA, Nagorney DM, Schroeder G, Silverstein MN. Splenectomy in myelofibrosis with myeloid metaplasia: a single-institution experience with 223 patients. *Blood*. 2000;95:2226-2233.
- Porcu P, Neiman RS, Orazi A. Splenectomy in agnogenic myeloid metaplasia [letter]. *Blood*. 1999;93:2132-2134.
- Kaplan EL, Meier P. Nonparametric estimation from incomplete observations. *J Am Stat Assoc*. 1958;53:457-481.
- Wolf BC, Banks PM, Mann RB, Neiman RS. Splenic hematopoiesis in polycythemia vera: a morphologic and immunohistologic study. *Am J Clin Pathol*. 1988;89:69-75.
- Dupriez B, Morel P, Demory JL, et al. Prognostic factors in agnogenic myeloid metaplasia: a report on 195 cases with a new scoring system. *Blood*. 1996;88:1013-1018.
- Wolf BC, Neiman RS. Hypothesis: splenic filtration and the pathogenesis of extramedullary hematopoiesis in agnogenic myeloid metaplasia. *Hematol Pathol*. 1987;1:77-80.
- Litam PP, Friedman HD, Loughran TP Jr. Splenic extramedullary hematopoiesis in a patient receiving intermittently administered granulocyte colony-stimulating factor. *Ann Intern Med*. 1993;118:954-955.
- O'Keane JC, Wolf BC, Neiman RS. The pathogenesis of splenic extramedullary hematopoiesis in metastatic carcinoma. *Cancer*. 1989;63:1539-1543.
- de Haan G, Dontje B, Engel C, Loeffler M, Nijhof W. The kinetics of murine hematopoietic stem cells in vivo in response to prolonged increased mature blood cell production induced by granulocyte colony-stimulating factor. *Blood*. 1995;86:2986-2992.
- Hibbin JA, Njoku OS, Matutes E, Lewis SM, Goldman JM. Myeloid progenitor cells in the circulation of patients with myelofibrosis and other myeloproliferative disorders. *Br J Haematol*. 1984;57:495-503.