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Osteomyelofibrosis/-sclerosis: A Histological and Cytogenetic Study on Core Biopsies of the Bone Marrow *

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Summary. A combined histological and cytogenetic study was performed on the bone marrow in 33 patients with overt osteomyelofibrosis/-sclerosis (MF/OMS) and so called agnogenic myeloid metaplasia including blast crisis.

Histopathology of the plastic embedded samples of bone marrow showed an abnormal proliferation of megakaryopoiesis with conspicuous atypias of growth and maturation in addition to a neoplastic neutrophilic granulopoiesis, particularly in the early stages of MF. Thus a biphasic population of neoplastic hematopoiesis is postulated and this lesion is called chronic megakaryocytic-granulocytic myelosis (CMGM) with myelofibrosis – CMGM stage III – or with osteomyelosclerosis – CMGM stage IV. Initiation of fibrillogenesis, the most striking alteration of this disorder, is partially attributed to disorganization of megakaryopoiesis with abnormal proliferation and clustering around the sinuses and intraluminal growth, with subsequent obliteration of the vascular compartment.

Cytogenetic evaluation demonstrated the Philadelphia chromosome (Ph'-chromosome) in 93% of CGL and in 67% of MF/OMS, including cases with blast crisis. Unlike CGL and MF/OMS where a Ph'-chromosome is common, myelofibrosis of non-neoplastic origin and AML displayed no Ph'-chromosome. Further aberrations such as aneuploidy involved the C/D group chromosomes predominantly and were especially prominent in blast crisis (about 50%) with no significant differences in CGL and MF/OMS or in AML. Our results of chromosomal analysis, evaluated in close context with histopathology, show no fundamental differences between CGL and myeloproliferative disorders of mixed cellularity, i.e., chronic megakaryocytic-granulocytic myelosis (CMGM). For this reason the terminal stages of fibrotic and osteosclerotic lesions belong into these categories of CMGM or CGL respectively. In conclusion MF/OMS are final stages or subtypes of CML,

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carrying the same chromosomal marker and demonstrating remarkable atypias of the hematopoietic tissue suggestive of malignancy. The fibrotic/osteosclerotic alteration itself is thought to represent a secondary non-neoplastic feature.

Key words: Myelofibrosis – Osteomyelosclerosis – Histopathology – Cytogenetics – Bone marrow biopsy.

Introduction

Osteomyelofibrosis/-sclerosis (MF/OMS) with myeloid metaplasia has widely been assumed to be one of the main features of hematological diseases collectively termed the myeloproliferative disorders (Dameshek 1951). It has further been proposed that MF/OMS and chronic myelogenous leukemia (CML) are closely related or even identical processes (reviews by Block et al. 1975; Georgii 1979). However, this latter concept which implies that MF/OMS is a secondary or accompanying feature in the course of CML is not accepted unequivocally by other hematologists (review by Bearman et al. 1979). To elucidate the true nature of this disorder morphological investigations should include both the study of chromosomal anomalies and the histopathology of the bone marrow. The demonstration of the Philadelphia (Ph') chromosome is particularly important since it is well established that this marker characterizes CML and is virtually diagnostic for that particular entity (Rowley 1976, 1979). In contrast with acute and chronic myeloid leukemia cytogenetic investigations have only rarely been performed in MF/OMS (review by Whang-Peng et al. 1978). This largely depends on technical factors, i.e., the fibrotic alteration of the bone marrow and the absence of cellular hematopoietic tissue. Consequently comprehensive chromosomal studies are only possible on direct preparations from marrow specimens obtained by trephine or core biopsies. They should not be restricted to peripheral blood cultures which have occasionally been reported (Nowell et al. 1976; Nowell and Finan 1978; Den Ottolander et al. 1979; Geraedts et al. 1980). A further disadvantage in evaluating chromosomal analysis in MF/OMS is the lack of a clear cut definition of various myeloproliferative diseases where overlaps occur, as stated by Bearman et al. (1979). Our definition of MF/OMS is based on clinical parameters and histopathology of the bone marrow, including sequential examination and autopsy findings. According to this there are two principle types of MF/OMS: *firstly* the idiopathic or primary MF of unknown cause (immunologic, inflammatory); *secondly* the MF/OMS with myeloid metaplasia arising from previous leukemic diseases or Polycythemia vera. In the majority of cases this secondary MF/OMS is the sequel of a clinically a- or subleukemic myelosis of mixed cellularity which is called chronic megakaryocytic-granulocytic myelosis (CMGM, see also Georgii 1979). However, the so called acute (malignant) MF/OMS (Lewis and Szur 1963) presents a blast crisis in a previously undiagnosed CML/CMGM rather than a distinct clinicopathological entity. This statement is supported by reviews of the current literature, especially by the variable finding of splenomegaly and the variable duration of the diseases (Buyssens and Bourgeois 1977; Bearman et al. 1979; Cheng

1979). Because of the fibrotic change of the bone marrow conventional puncture and aspiration procedure is often unsuccessful (so called dry tap) and a biopsy becomes essential. It is also necessary for a thorough histological examination and an elaborate technique of processing marrow samples is required to detect and evaluate the cellular compositions of MF or OMS.

In this study the findings of histopathology and cytogenetics in 33 patients with overt MF/OMS partially evolving into a blast crisis are reported and compared with chronic granulocytic leukemia (CGL). In addition 14 patients with acute myeloid leukemia (AML) and 1 case with idiopathic (primary) myelofibrosis were investigated as controls.

Material and Methods

Patients. A total of 275 patients with MF/OMS were examined and the final diagnosis established by histology of the bone marrow specimen in correlation with clinical findings. Of these 8 turned out to evolve into MF/OMS as a sequel of a long standing Polycythemia vera (P. vera) and 15 patients presented a primary (idiopathic) myelofibrosis of unknown cause. The remaining 252 patients displayed the clinical picture of a myeloproliferative disorder with or without blast crisis or acute onset of disease and with varying degrees of MF/OMS, mostly accompanied by so called agnogenic myeloid metaplasia. In only a fraction of those patients (33 cases) with MF/OMS were simultaneous histological and cytogenetical investigations possible. The 8 cases with MF in the course of P. vera have been reported elsewhere and are not included in this study (for more details see Vykoupil et al. 1980). For controls, chromosomal analysis was also performed in one case of idiopathic myelofibrosis, in 14 patients with overt AML and in 41 cases with CGL.

Methods. The procedure for trephine biopsies of the bone marrow followed the methods described by Burkhardt (1966) or Jamshidi and Swaim (1971). For *cytogenetic investigation* samples of marrow were obtained by cutting off tiny pieces from the core at its medial (innermost) part and extruding the marrow from the spaces between the osseous trabecula.

By this procedure it was possible to extract sufficient material in most cases with severe OMS. Metaphases were obtained by direct bone marrow preparations without phytohemagglutinin (PHA) stimulation and from short-time cultures together with the simultaneously drawn blood sample. Staining was performed by conventional methods and the Giemsa-banding technique and chromosomal analysis was done on photographs. For *light microscopy* the remaining two thirds of the core of bone marrow were fixed, embedded in plastic (methylmethacrylate) and further processing was done with semithin sections and staining procedures (Giemsa, Goldner's trichrome, silverimpregnation after Gomori, Prussian blue reaction and methylgreen-pyronin) following the methods described by Burkhardt (1971).

Results

The *histopathology* of MF/OMS is characterized by an overt increase of reticulin and collagen fibers with a felt-like appearance and partial obliteration of the marrow spaces by striking apposition of newly formed bone (Figs. 1a, b, 2a, b). Besides these conspicuous alterations of the myeloid stroma and the osseous trabeculae there is a varying population of mostly bizarre hematopoietic cells entrapped in the dense reticulum of fibers and endophytic bone (Figs. 1a, b – 3a, b). A closer view on the lesions of the bone marrow in MF displays a remarkable growth of megakaryocytes forming small clusters in this network of thickened reticulin and small bundles of collagen fibers. Those fibers are easily visible in silver impregnated specimens and the former are even recognizable without polarization (Fig. 1a). In the majority of cases the megakaryocytes

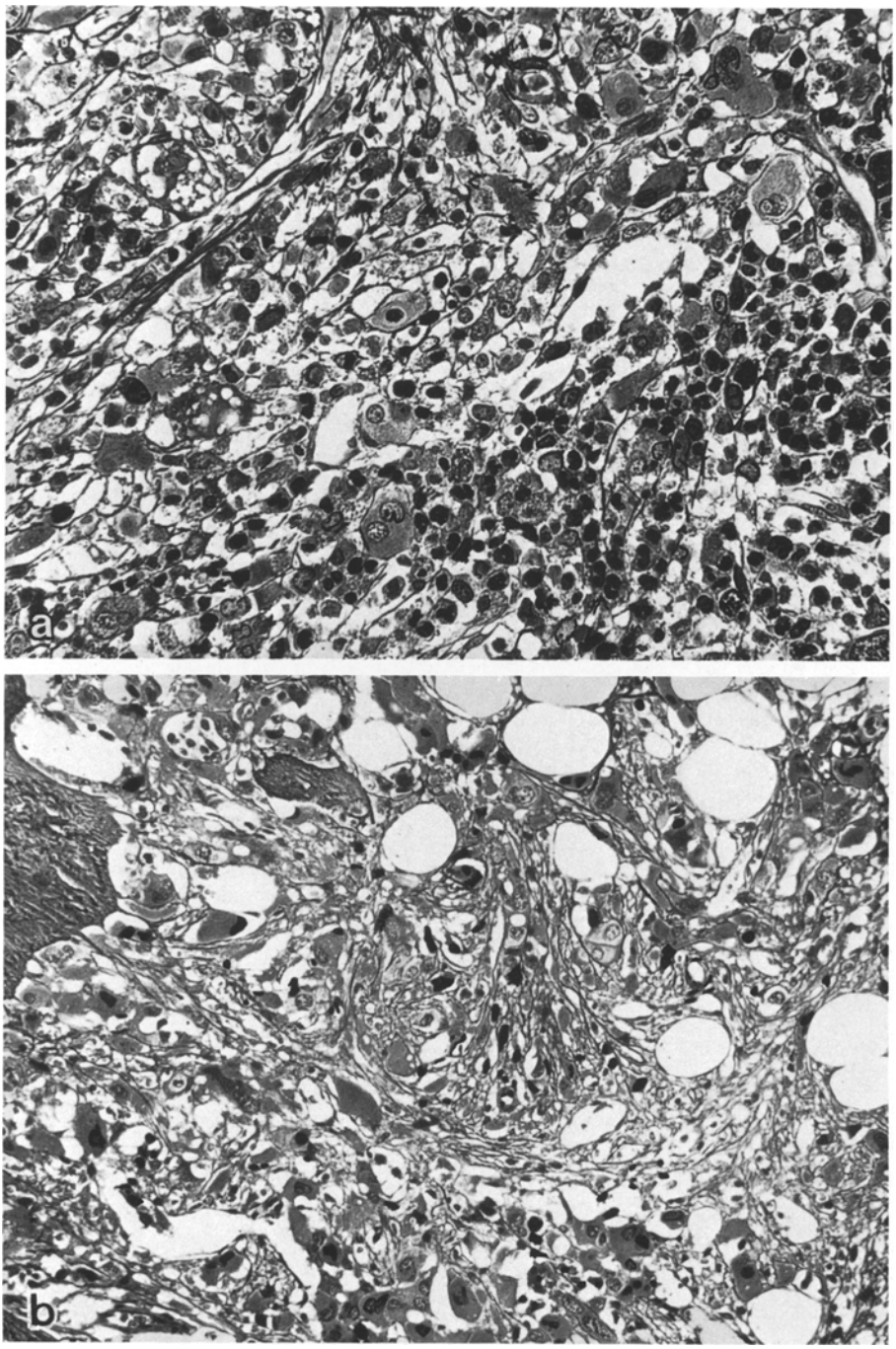


Fig. 1a, b. Survey of myelofibrosis – osteomyelosclerosis. **a** Myelofibrosis (MF) with a conspicuous growth of megakaryocytes and precursors of segmented granulocytes in a network of reticulin fibers and small bundles of collagen fibrils (*left corner*). This lesion corresponds to chronic megakaryocytic-granulocytic myelosis (CMGM) stage III. **b** Osteomyelosclerosis (OMS) with endophytic bone formation (*left corner*) and a dense mesh of collagen fibers entrapping polymorphous megakaryocytes. This lesion is consistent with CMGM stage IV (see text). **a** × 280 – Gomori; **b** × 210 – Gomori

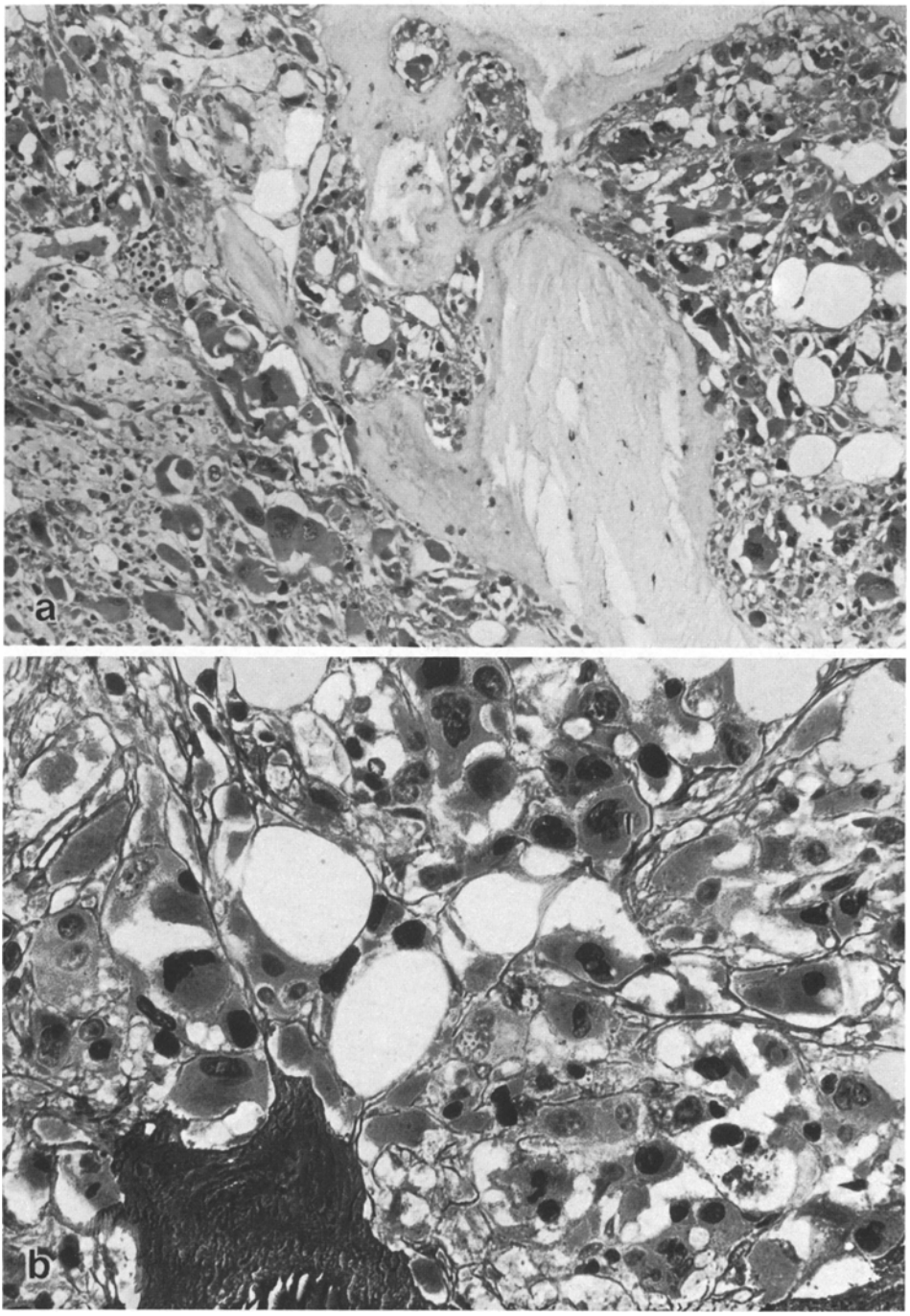


Fig. 2a, b. Osteomyelosclerosis (OMS or CMGM stage IV). **a** Partial obliteration of the marrow space by extensive growth of endophytic bone surrounded by reticulin and collagen fibers, which enclose clusters of bizarre megakaryocytes and residues of granulo- and erythropoiesis. **b** Collagen fibrils with assemblies of megakaryocytes which display polymorphism with a variety of nuclear aspects and sizes, in close connection with newly formed bone (*lower half*). **a** × 180 – Giemsa; **b** × 320 – Giemsa

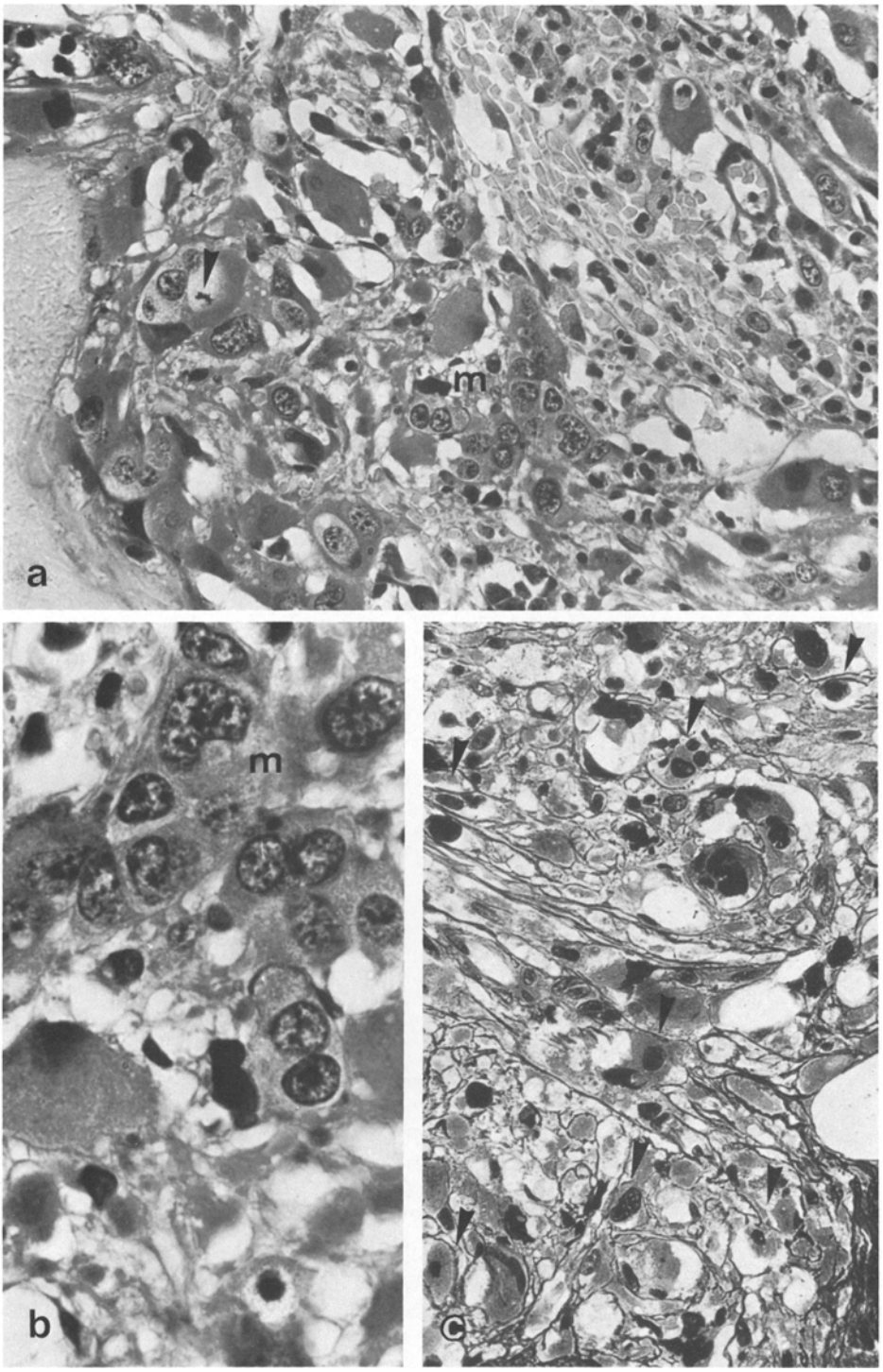
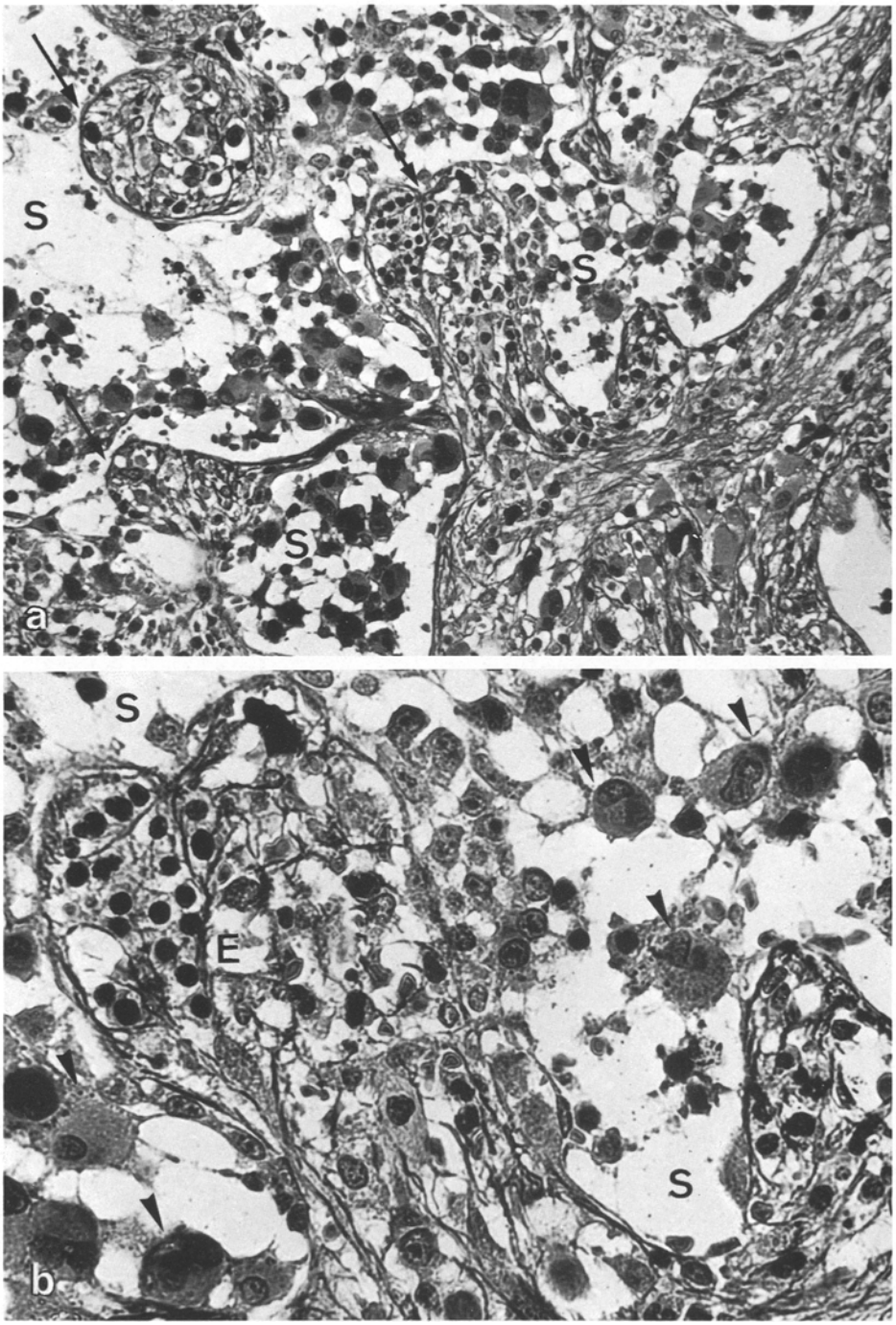


Fig. 3a–c. Atypia of megakaryopoiesis in MF/OMS – abnormal cytological differentiation. **a** Survey with cluster of bizarre megakaryocytes dislocated towards the peritrabecular generation zone of neutrophilic granulopoiesis and showing a mitosis (*arrow head*) and many immature and precursor forms (*m*). **b** Grouping a megakaryoblasts and very early megakaryocytes with large indented or bilobated nuclei (*m*). **c** Micromegakaryocytes lying along collagen fibrils in OMS and displaying chromatin dense or pyknotic nuclei with only a few segments (*arrow heads*). **a** × 320 – Giemsa; **b** × 780 – Giemsa; **c** × 320 – Gomori

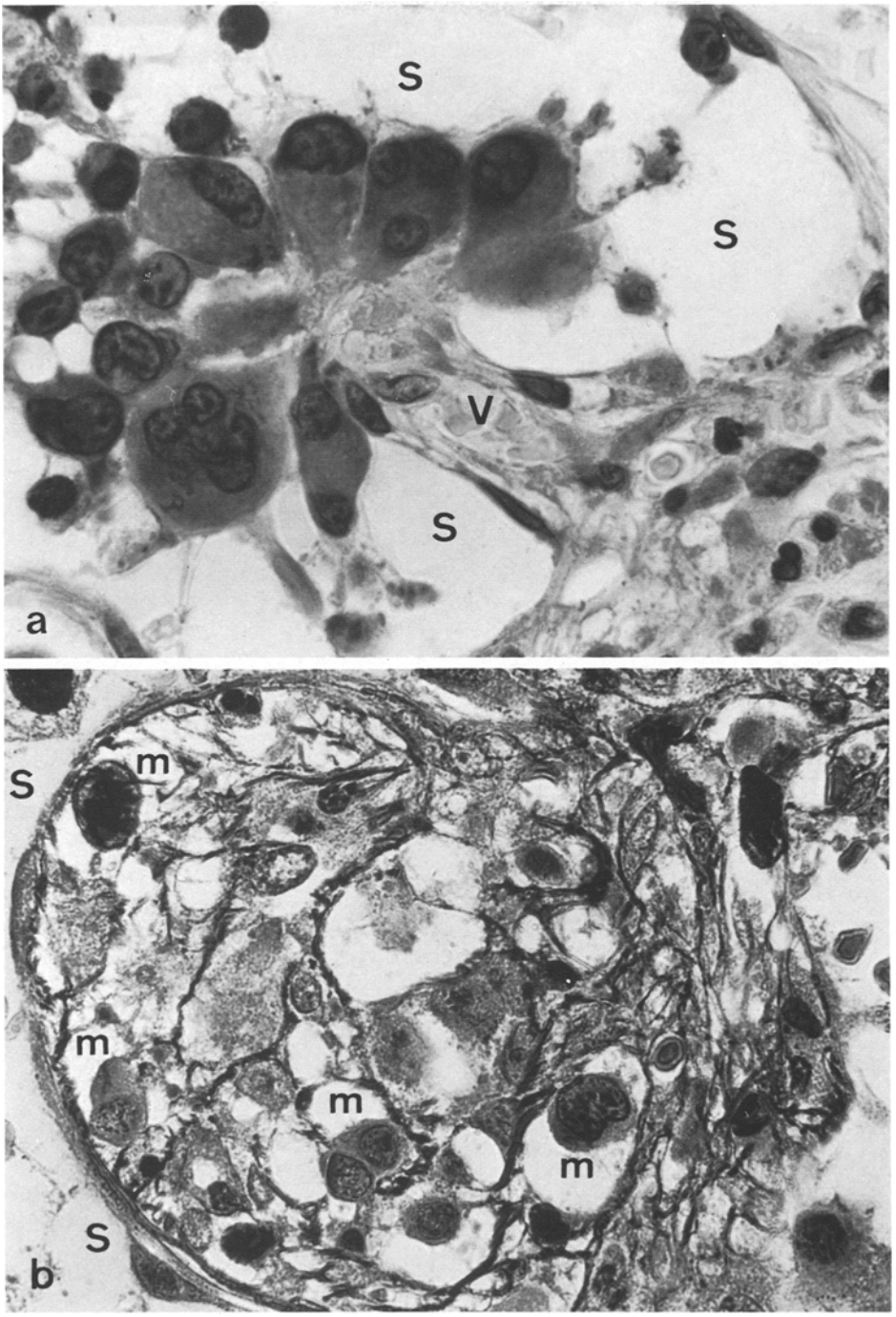
are small and comparable with those of a more advanced stage of MF with newly formed bone and broad bundles of collagen fibrils, i.e. OMS (Figs. 1b, 2a, b). A more detailed description of the megakaryocyte fine structure includes an apparent disturbance of maturation apart from the variety of forms and the presence of many so called micromegakaryocytes. There is a frequent assembly of megakaryopoiesis along the osseous trabecula which is the original generation zone of granulopoiesis (Figs. 2a, b, 3a) an increased ratio of mitoses (Fig. 3a) and a remarkable shift to the left with many immature and precursor forms, hardly recognizable as belonging to the megakaryocyte lineage (Fig. 3a, b). Large nuclei with prominent nucleoli and a flocculent dispersion of chromatin may be observed, accompanied by shallow indentations or bilobation of the nuclei (Fig. 3b). There are also numerous micromegakaryocytes which are difficult to detect among the other cells between the bundles of reticulin fibers, including pyknotic forms and so called naked nuclei (Figs. 2b, 3c). This polymorphism of megakaryopoiesis is further augmented by several oversized or giant forms with are most conspicuous in surveys (Fig. 2a). There is an enormous clustering of megakaryocytes arround the venous sinusoids in addition to cytological atypia and disorganization of maturation (Figs. 1a, 4a). This clustering progresses into a striking prolapse or herniation of proliferating megakaryocytes into the lumina of the sinuses (Figs. 4a, b, 5a). A remarkable dilatation and increase in number of these sinuses with their branched vascular channels is frequently accompanied by an intraluminal megakaryopoiesis which produces an extensive grouping of giant- and microforms (Figs. 4a, b) besides bulging islets of erythropoiesis (Figs. 4b). These toadstool-like protuberances of megakaryopoiesis may not only create a bizarre deformation of the sinuses but probably causes partial obliteration of the lumina. In addition megakaryopoiesis is providing abnormal microforms for shedding into the lumina (Figs. 5a, b) with so called dissociation of nuclear/cytoplasmic maturation as mentioned above.

Although in MF/OMS megakaryopoiesis may be most conspicuous, the other cell lineages also display certain abnormalities of proliferation. In MF especially neutrophilic granulopoiesis and erythropoiesis may be prominent and show many precursor forms, but in the chronic stages of this disease these display a full maturation to polymorphonuclear cells or erythrocytes respectively (Figs. 1a, 2a, 3a). In contrast the acute phase of MF/OMS or so called malignant osteomyelofibrosis/-sclerosis is characterized by an increase of immature or progenitor cells of both megakaryo- and granulopoiesis. In a manner similar to an incipient blast crisis in CGL (Fig. 6a) where myeloblast proliferation starts at the peritrabecular generation zones with a lack of further differentiation (the so called hiatus leukaemicus) a comparable feature may be observed in MF/OMS. Atypical cellular proliferation and failure to differentiate often commences at the osseous trabecula. Here a dense infiltrate of bizarre micromegakaryocytes, consistent with so called acute megakaryocytic myelofibrosis, is visible (Fig. 6b) alternatively, a population of myeloblasts lying between the collagen fibers with an identical cytology to AML or to blasts in an acute transformation of CGL (Fig. 6c) is seen.

The different appearences of MF/OMS in acute or terminal phases of this disease have to be clearly separated from an entity called idiopathic or primary



Figs. 4a, b–5a, b. Atypia of megakaryopoiesis in MF/OMS – abnormal proliferation. **Fig. 4. a** Survey with dilated and branched sinus displaying an irregular outline due to herniation of proliferating megakaryopoiesis and erythropoiesis (*arrows*). In the sinus (*S*) intraluminal growth and clustering of megakaryocytes, mostly microforms. **b** High magnification of bulging cluster of erythropoiesis (*E*) containing normoblasts with dense round nuclei. Intrasinusoidally (*S*) there are many microme-



gakaryocytes (*arrow heads*). **Fig. 5. a** Toadstool-like proliferation of megakaryopoiesis with herniation into a sinus lumen (*S*). Megakaryocytes display a varity of differentiation mostly into microforms. These desquamate into the lumen of the venous sinusoid with a small vessel (*V*) at the base of this grouping. **b** Bleb-like protuberance of micromegakaryocytes (*m*) into a sinus (*S*) with surrounding reticulin fibers. **4a** × 260 – Gomori; **4b, 5b** × 780 – Gomori; **5a** × 780 – Giemsa

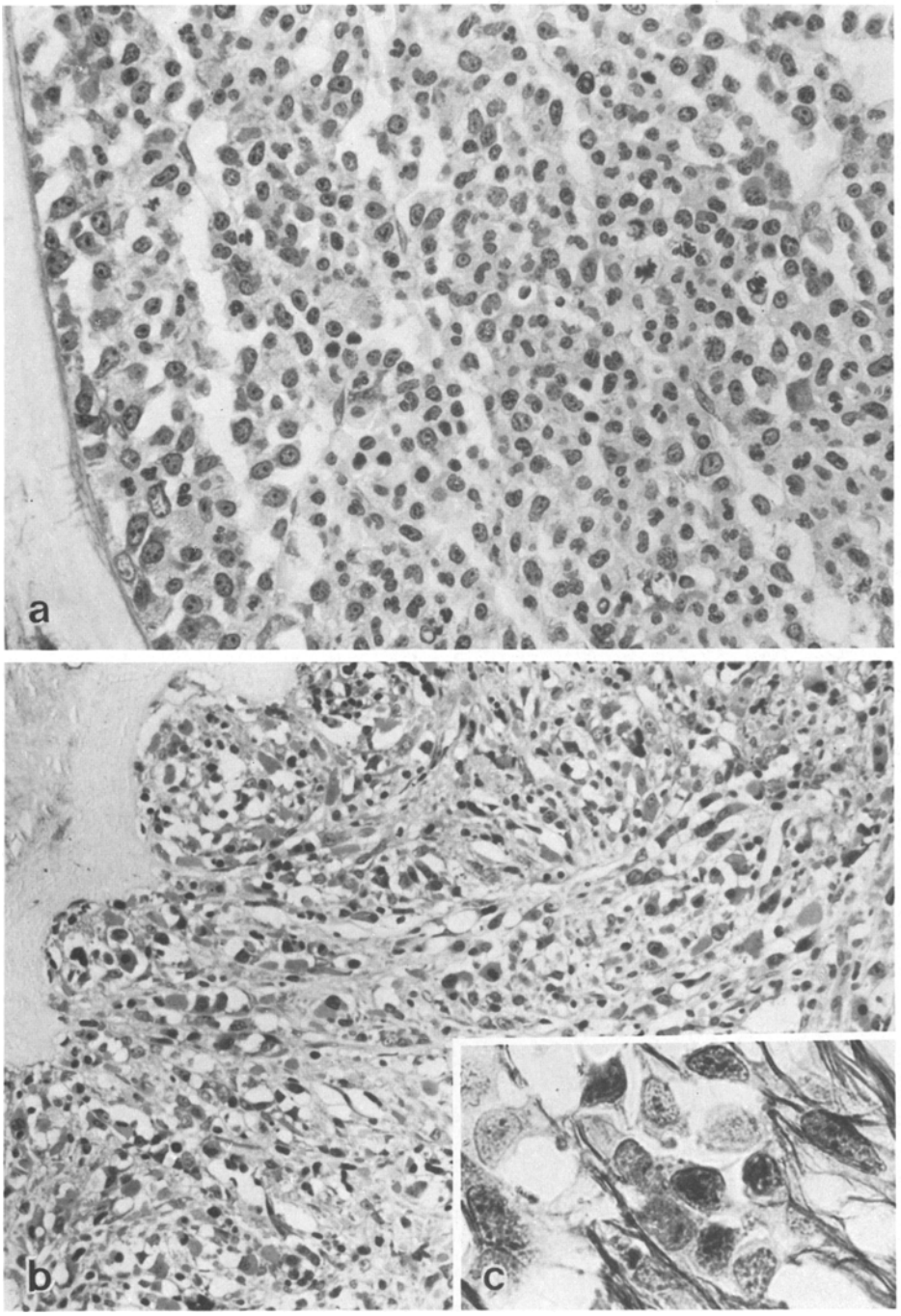


Fig. 6a–c. Blast crisis in OMS in comparison with chronic granulocytic leukemia (CGL). **a** CGL with conspicuous increase of blasts, proliferating at the peritrabecular generation zone with progression into the center of the marrow space. This contains only few segmented neutrophils (so called hiatus leukaemicus). **b** OMS with abnormal proliferation of numerous micromegakaryocytes apparently originating from the peritrabecular zone of endophytic bone and progressing towards the marrow center, surrounded by reticulin and collagen fibers – so called micromegakaryoblastic crisis (acute or malignant myelofibrosis/osteomyelosclerosis). **c** Blastic crisis in OMS with probably myeloblasts between collagen fibrils, without further differentiation. **a** × 320 – Giemsa; **b** × 200 – Giemsa; **c** × 780 – Gomori

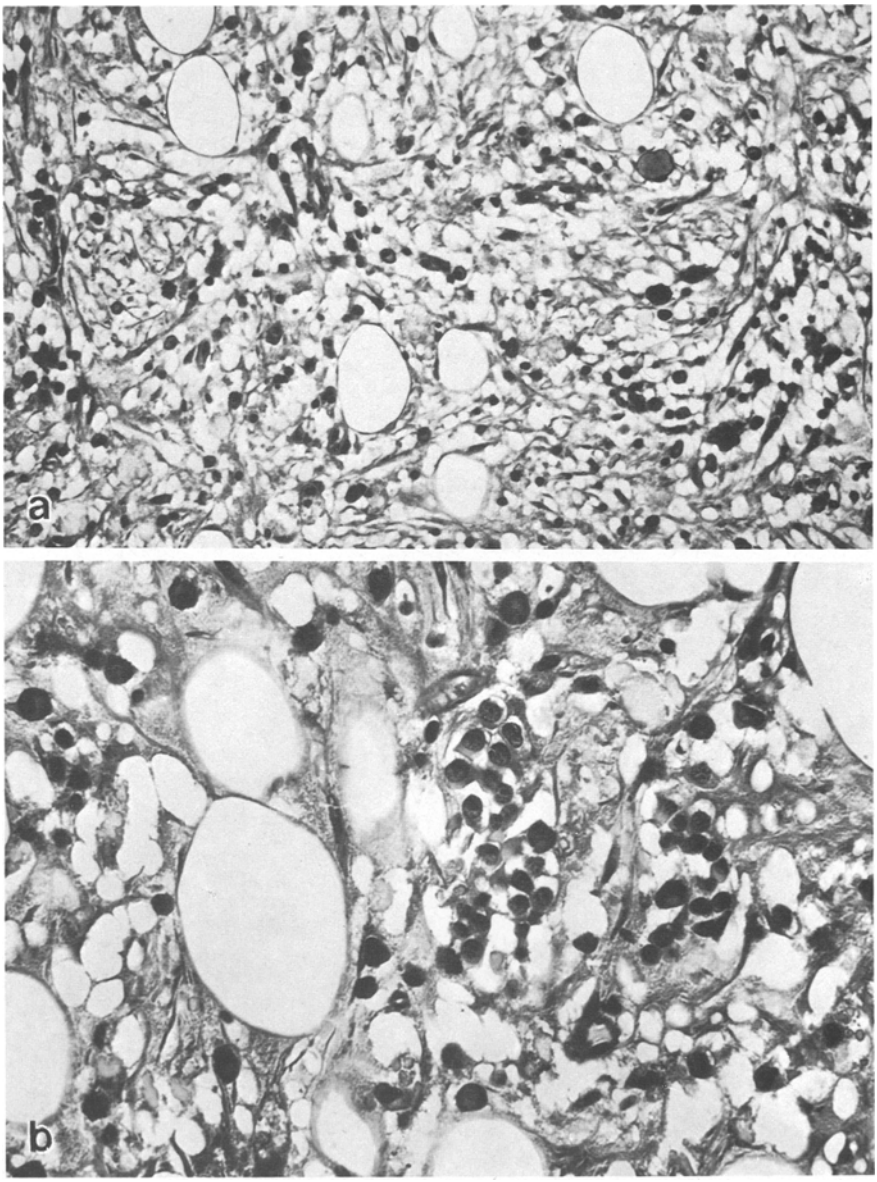


Fig. 7a, b. Primary (idiopathic) myelofibrosis of unknown cause. **a** Survey with scleredema of the marrow with meshwork of reticulin fibers entrapping mainly mononuclear cells. **b** High magnification shows clusters of plasma cells, but no atypical or increased proliferation of megakaryocytes or neutrophilic granulocytes. **a** × 160 – Giemsa; **b** × 340 – Giemsa

myelofibrosis. In these cases the bone marrow shows a scleredema with reticulin fibers but only single strands of collagen and a cellular infiltrate consisting predominantly of plasma and mast cells (Fig. 7a, b). In contrast to MF/OMS there is a partial aplasia of hematopoiesis, but no significant proliferation or atypia of the megakaryo-granulopoiesis is detectable (Fig. 7b).

Cytogenetic analysis of these different chronic or acute stages of MF/OMS is given in Table 1 as a survey with an emphasis on the presence of the Ph'-chromosome and aneuploidy with anomalies of C and D group chromosomes. Table 2 shows a comparison of karyotypes in CGL and overt MF/OMS (CMGM III, IV). From these results it is obvious that the marker chromosome (Ph') may be detected in the bone marrow of MF/OMS in a frequency which is only 26% lower than in CGL or CML respectively. A similar finding is encountered when examining aneuploidy of the C and D group chromosomes where no significant difference of those non-random changes of the karyogram exists between these two entities (MF/OMS versus CGL/CML). In addition to this overview Table 1 further demonstrates that chromosomal aberrations were observed in the C and D group in 3 cases of Ph'-negative CGL including blast crisis (+9, +15) and also in those cases of AML. MF/OMS with a blast crisis (malignant or acute osteomyelosclerosis, 11 cases) had the highest ratio of clonal evolution and abnormalities of the C- and D-group chromosomes. The frequency of these abnormalities was comparable only with CGL or AML. The one case with primary (idiopathic) MF of apparently non-neoplastic origin displayed no major chromosomal anomalies.

Table 1. Survey of cytogenetic findings in 74 patients with a chronic myeloproliferative disease in comparison with acute myeloid leukemia (14 patients)

Histopathology	Number of patients (*n*)	Ph'-positive		Clonal evolution of Ph'		Aneuploidy				Mean of metaphases examined
		Ratio	Percentage	Ratio	Percentage	Total		C/D-group		
						Ratio	Percentage	Ratio	Percentage	
Chronic granulocytic leukemia (CGL)	27	25/27	93	7/27	26	15/27	56	11/27	41	16
with blast crisis (*control*)	14	13/14	93	3/14	21	8/14	57	7/14	50	18
Osteomyelofibrosis/-sclerosis (i.e., myelosclerosis with myeloid metaplasia)	22	14/22	64	4/22	18	10/22	45	4/22	18	12
with blast crisis (i.e., acute myelofibrosis/malignant osteomyelosclerosis)	11	8/11	73	7/11	64	9/11	82	6/11	55	15
Acute myeloid leukemia (AML) (*control*)	14	0/14	–	–	–	7/14	50	4/14	29	17

Table 2. Comparison of cytogenetic findings in chronic granulocytic leukemia (CGL) including blast crisis with overt osteomyelofibrosis/-sclerosis, i.e., myelosclerosis with myeloid metaplasia, also partially with blast crisis (so called CMGM III, IV)

Histopathology	Number of patients (*n*)	Ph'-positive (Percentage)	Aneuploidy			
			Total		C/D-group	
			Ratio	Percentage	Ratio	Percentage
Chronic granulocytic leukemia (CGL) including blast crisis	41	93	23/41	56	18/41	44
Overt osteomyelofibrosis/ -sclerosis including blast crisis	33	67	19/33	58	10/33	30
Total	74	81	42/84	57	28/74	39

Discussion

A critical evaluation of our results should be focussed on three major points:

1. Findings suggestive for a neoplastic or malignant nature of MF/OMS.
2. Origin and development of the striking fibrotic alterations of the bone marrow.
3. Chromosomal anomalies in comparison with the karyograms in CML.

Since the reports of Bouroncle and Doan (1962) and Lewis and Szur (1963) on malignant myelosclerosis or myelofibrosis there have existed two differing opinions on the true nature of this disease, which is counted among the so called myeloproliferative syndromes (Dameshek 1951). It may represent a distinct entity or a peculiar form of leukemia/myelosis. Clinical and morphological findings point towards a malignant or neoplastic origin when reviewing larger series (Gralnick et al. 1971) or individual more recently published case reports (Estevez et al. 1974; Lubin et al. 1976; Fabich and Raich 1977; Bird and Proctor 1977; Cheng 1979; Bearman et al. 1979). This presumed malignant origin is also suggested by the conspicuous cytological atypia of the proliferating cell lineages, predominantly megakaryopoiesis. In addition to the often bizarre appearance and disorganization of maturation which is documented in the various illustrations of several papers (Buyssens and Bourgeois 1977; Bird and Proctor 1977; Fabich and Raich 1977; Bearman et al. 1979) and extended by our findings, there are also abnormalities of histochemical reactions (Lobdell and Europa 1962). Ultrastructural alteration, especially in cases with evolution into a blastic crisis, are further arguments for an underlying malignancy (Thiele et al. 1980). This is further supported by the clinically observed acute onset of this disease or of blastic transformation itself (review by Pedersen 1973) which is compatible with so called acute (malignant) myelosclerosis (Lewis and Szur 1963). In this context of assumed malignancy the different populations of proliferating cell lines have to be considered, since there is also an apparently neoplastic granulopoiesis apart from the megakaryopoiesis. In the later stages of MF particularly, this granulopoiesis may decrease and become inconspicuous

as an overwhelming fibro- and osteosclerosis develops. These terminal presentations of OMS could be called "burnt out" stages of the disease. However, a biphasic or bivalent neoplasia consisting of neutrophils and megakaryocytes or a mixed cellularity can be demonstrated in the earlier phases of this disorder, prior to severe scarring. Between those early stages of MF with only slight to moderate fibrosis and increased cellular proliferation, and the so called myelosis of mixed cellularity or CMGM (Georgii et al. 1980) the differences are only gradual. Consequently it is worthwhile speculating that the stages of incipient or minimal fibrosis – CMGM II – with demonstration of reticulin fibers after polarization of silver impregnated specimens, present the immediate precursors of overt MF and OMS respectively. This hypothesis can be further discussed in context with the chromosomal findings.

Evolution of myelofibrosis is assumed to occur step-wise with steps of unknown duration. Only clinical observations with a close follow up of the course of disease, including sequential core biopsies of the bone marrow, may elucidate this process. However, until now only a very few patients have been studied by such repeated examination (Thiele et al. 1980; Vykoupil et al. 1980). Atypical proliferation and disturbed maturation of megakaryocytes may play an important role in fibrillogenesis. A fibrotic reaction of the mesenchymal compartment of the bone marrow may be caused or at least initiated by an abnormal release of biogenic amines (serotonine, histamine) from the specific granules of fragmented megakaryocytes and platelets (Zucker-Franklin 1975; Thiele et al. 1977, 1980). Another possibility which is suggested by the histological findings concerns the partial obliteration of the venous sinusoids. The atypical growth of megakaryopoiesis around the sinuses with destruction of vascular walls and intraluminal protrusion (Figs. 4 and 5) may cause disorganization of the microenvironment with resultant ischemia and scarring of the myeloid stroma, as proposed by Georgii and Vykoupil (1976). This would imply a scirrhous-like change of the mesenchyme comparable with solid tumors (Lennert et al. 1964; Georgii and Vykoupil 1976). The fibroblasts involved in this process of myeloid scarring, do not display the Ph'-chromosome or other major chromosomal anomalies as reported in CML with or without MF (Maniatis et al. 1969; Van Slyck et al. 1970; De la Chapelle et al. 1973). Consequently fibrillogenesis seems to be a concomitant or secondary feature which accompanies malignant hematopoiesis.

Cytogenetic investigations demonstrate that a *Ph'-chromosome* is present in 67%, i.e., 22 out of 33 MF/OMS cases examined including the terminal or blastic phase of this disease. This high incidence may be compared with the frequency of the marker chromosome in CGL, where in 38 out of 41 (93%) a Ph'-chromosome was detected. This difference of occurrence of the Ph'-chromosome in MF/OMS (67%) and CGL (93%) should be related firstly to the difficulty to obtain sufficient cells from the scarred marrow and secondly to genuinely Ph'-chromosome negative cases.

In MF/OMS the presence of a Ph'-chromosome has been recorded only occasionally but in more than one patient by Krauss (1966, 5 cases) and Müller and Haberlandt (1970, 3 cases) and in individual case reports (Forrester and

Louro 1966; Cohen 1967; Frey and Siebner 1968; Khan and Martin 1968; Rosenthal and Moloney 1977). Two larger series and reviews (Whang-Peng et al. 1978, 20 cases) and Rowley (1977, 7 cases) show non-random changes of chromosomes in MF/OMS only but as already mentioned this may have several explanations including heterogeneity of the clinical parameters and laboratory data studied in patients suspected of suffering with MF/OMS, without establishment of an exact diagnosis by histopathology of trephine biopsies of the bone marrow. Moreover, there are the difficulties of obtaining a sufficient marrow sample particularly in cases of advanced OMS and the restriction of karyotyping cells from peripheral blood cultures (Nowell et al. 1976; Den Ottolander et al. 1979; Geraedts et al. 1980). The occurrence of the marker chromosome in MF/OMS is further confirmed by the fact that our classification (Georgii 1979; Georgii et al. 1980) includes subtypes which are thought to present some kind of transition between CML and overt MF/OMS and which are called CMGM I, II (agnogenic myeloid metaplasia without or with minimal fibrosis). It is suggested that these subtypes present forms of transgression towards MF and show a similar incidence of Ph'-chromosome positive cases and other non-random aberrations as overt MF/OMS (Georgii et al. 1980). In addition to these results providing evidence of a clonal chromosomal marker in MF/OMS, a clonal proliferation of hematopoietic stem cells with MF was observed in a case of agnogenic myeloid metaplasia (corresponding to MF or CMGM III, see below) by demonstration of the enzymatic marker glucose-6-phosphate-dehydrogenase (Jacobson et al. 1978). This clearly supports our concept of a clonal and neoplastic proliferation in MF/OMS which may be called CMGM stage III or IV when referring to the fibrotic/osteosclerotic lesions of the marrow.

Non-random anomalies of chromosomes in MF/OMS have been found in about 58% of cases which correlates with the results (48–65%) published by Whang-Peng et al. (1978) but not with the 15% reported by Rowley (1977). These anomalies are comparable to similar aberrations in CGL with and without blastic crisis, in our series and in others (reviews by Rowley 1978; Van den Berghe et al. 1978). Those chromosomal abnormalities are of a complex and non-specific nature and present a common finding in MF/OMS. They most frequently involve the C- and D-groups with appearance of a monosomy 7 and trisomy 8 or 9 (Sandberg et al. 1964; Kiossoglou et al. 1966; Jackson and Higgins 1967; Mitus et al. 1969; Nowell et al. 1976; Rowley 1977; Jacobson et al. 1978; Nowell and Finan 1978). This involvement of C-D chromosomes is almost identical with CML in the chronic or blastic stage (Gahrton et al. 1974; Rowley 1978, 1979). As Whang-Peng et al. (1978) stated, these cytogenetic data of non-random aberrations reflect the relationship which exists between leukemias and the various myeloproliferative diseases and MF/OMS. The frequent occurrence of the Ph'-chromosome however, characterizes MF/OMS as a subtype of CML and therefore extends and confirms earlier findings of histopathology, particularly regarding the so called myeloses of mixed cellularity or CMGM (Georgii et al. 1980).

Histopathology – including cellular composition, atypicality and the gradually occurring process of fibrosis – and the cytogenetic findings of a frequent

Ph'-chromosome marker, support our concept that MF/OMS may be called an advanced stage of CMGM with overt fibrotic or/and osteosclerotic alterations of the bone marrow, or CMGM stage III (MF), and CMGM stage IV (OMS).

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