

Chronic Idiopathic Myelofibrosis

John T. Reilly

Contents

15.1	Introduction	254
15.2	Pathogenesis	254
15.2.1	Clonality	254
15.2.2	Cytogenetics	255
15.2.3	Molecular Studies	256
15.2.4	Role of Growth Factors	256
15.2.4.1	Platelet-Derived Growth Factor	256
15.2.4.2	Transforming Growth Factor- β	257
15.2.4.3	Additional Growth Factors and Cytokines	258
15.2.5	Animal Models	259
15.3	Diagnosis	259
15.4	Clinical Manifestations	261
15.5	Laboratory Features	264
15.6	Prognosis	264
15.7	Management	266
15.7.1	Medical Therapy	266
15.7.1.1	Cytotoxic Therapy	266
15.7.1.2	Androgens	266
15.7.1.3	Erythropoietin	266
15.7.1.4	Interferon	266
15.7.1.5	Thalidomide	267
15.7.1.6	Experimental Therapy	267
15.7.2	Surgery and Radiotherapy	267
15.7.2.1	Splenectomy	267
15.7.2.2	Radiotherapy	268

15.7.3	Stem Cell Transplantation	268
15.7.3.1	Standard Allo-SCT	268
15.7.3.2	Reduced Intensity Allo-SCT	269
15.7.3.3	Autologous SCT	269

References	269
------------	-----

Abstract. Chronic idiopathic myelofibrosis (CIMF) is a clinico-pathological entity characterized by a stem-cell-derived clonal myeloproliferation, extramedullary hematopoiesis, proliferation of bone marrow stromal components, splenomegaly, and ineffective erythropoiesis. It is the least common of the chronic myeloproliferative disorders and carries the worst prognosis with a median survival of only 4 years. Treatment for most cases is supportive, while androgens, recombinant erythropoietin, steroids and immuno-modulatory drugs are effective approaches for the management of anemia. Splenectomy and involved field irradiation may also be beneficial in carefully selected patients. Cure is only possible following bone marrow transplantation and a number of practical prognostic scores are available for identifying patients that would benefit from this approach. Recently, the use of low intensity conditioning has resulted in prolonged survival and lower transplant-related mortality. Finally, the recent reports of the association of CIMF with a gain-of-function JAK2 mutation opens the door to targeted therapies as well as molecular monitoring of treatment response.

15.1 Introduction

Chronic idiopathic myelofibrosis (CIMF), or myelofibrosis with myeloid metaplasia (MMM), is a chronic stem cell disorder characterized by bone marrow fibrosis, extramedullary hematopoiesis, splenomegaly, and a leuko-erythroblastic blood picture. It is an uncommon disorder, with a reported annual incidence ranging from 0.5 to 1.3 per 100,000 (Dougan et al. 1981; Mesa et al. 1997), with the highest rates being found among the Ashkenazi Jews in northern Israel (Chaïter et al. 1992). The etiology of CIMF is unknown, although environmental factors may be relevant as the disorder has been linked in a small number of patients to radiation (Andersen et al. 1964) and benzene exposure (Hu 1987). Although first described by Heuck in 1879, it was not until 1951, following Dameshek's seminal publication (Dameshek 1951), that the disease was regarded as one of the chronic myeloproliferative disorders. Recently, considerable progress has been made in understanding its pathogenesis, although this has yet to result in significant therapeutic advances. Indeed, its prognosis remains poor when compared to other *BCR-ABL*-negative chronic myeloproliferative disorders (Rozman et al. 1991), with death resulting from cardiac failure, infection, hemorrhage, and leukemic transformation.

15.2 Pathogenesis

15.2.1 Clonality

It has been appreciated for many years that CIMF is a clonal disorder and that the disease arises from the proliferation of malignant pluripotential stem cells. Such a conclusion was first suggested by early studies of the X-chromosome inactivation patterns of G-6-PD in patients who were heterozygous for this gene (Jacobson et al. 1978; Kahn et al. 1975). However, the low frequency of G-6-PD heterozygotes in the general population has led several groups to analyze the more informative X-linked genes, hypoxanthine phosphoribosyl transferase (HPRT) and phosphoglycerate kinase (PGK). In these studies, monoclonal hematopoiesis was documented in all patients irrespective of whether they had early cellular phase disease or more advanced myelofibrosis (Kreipe et al. 1991; Tsukamoto et al. 1994). Recently, Reeder and colleagues (2003), using fluorescent in situ hybridization (FISH), have provided evidence that both

B and T cells can be involved, while karyotypic analysis has shown that the stromal proliferation is polyclonal, or reactive, and not part of the underlying clonal hematopoiesis (Jacobson et al. 1978; Wang et al. 1992). Involvement of the B and T lymphocytic lineage was also suggested by an earlier study that utilized *N-Ras* gene mutational analysis, again supporting the pluripotent stem cell origin of the disease (Buschle et al. 1988). An increased number of circulating hematopoietic precursors, including pluripotent (CFU-GEMM) and lineage restricted progenitor cells (BFU-E, CFU-GM, and CFU-MK), is a feature of CIMF (Carlo-Stella et al. 1987; Han et al. 1988; Hibbin et al. 1984) and is likely to result from the proteolytic release of stem cells from the marrow (Zu et al. 2005). It is also possible that the spleen and liver contribute to the circulating progenitor pool (Wolf and Neiman 1987) as splenectomy temporarily normalizes levels (Craig et al. 1991). The high level of circulating progenitor cells is reflected in the significantly increased peripheral blood CD34⁺ cell count (Andreasson et al. 2002; Arora et al. 2004). Indeed, it has been proposed that not only can the absolute number of CD34⁺ cells be used to differentiate CIMF from other Philadelphia (Ph)-negative CMPDs, but the levels may also predict evolution to blast transformation (Barosi et al. 2001). Increased sensitivity of committed erythroid progenitors to erythropoietin has been reported (Carlo-Stella et al. 1987), while CFU-MK may exhibit autonomous growth (Han et al. 1988; Taksin et al. 1999) and/or hypersensitivity to interleukin-3 (Kobayashi et al. 1993). Such findings, coupled with the fact that autonomous megakaryocyte growth is not related to *MPL* mutations or autocrine stimulation by *Mpl-L* (Taksin et al. 1999), suggest that events downstream from receptor-ligand binding are likely to be pathogenetically important (Taksin et al. 1999). Finally, indirect evidence for the involvement of the pluripotential stem cell is provided by the rare reports of acquired hemoglobin H disease (Veer et al. 1979), paroxysmal nocturnal hemoglobinuria (Nakahata et al. 1993; Shaheen et al. 2005), acquired Pelger-Huet anomaly and neutrophil dysfunction (Perianin et al. 1984), as well as many abnormalities of platelet function (Cunietti et al. 1981; Schafer 1982).

15.2.2 Cytogenetics

Cytogenetic studies have played a pivotal role in the elucidation of pathogenetically important oncogenes in many hematological malignancies although, until recently, the data for CIMF has been sparse and confusing. However, over the last 15 years the publication of three large studies, involving a total of 256 well-characterized patients, has helped to clarify the situation (Demory et al. 1988; Reilly et al. 1997; Tefferi et al. 2001a). All three studies, as well as a literature review of 157 abnormal cases (Bench et al. 1998), have revealed that deletions of 13q and 20q, trisomy 8 and abnormalities of chromosomes 1, 7, and 9 constitute more than 80% of all chromosomal changes in CIMF. Deletions of 13q are the most common cytogenetic abnormality, occurring in approximately 25% of cases with an abnormal cytogenetic analysis (Demory et al. 1988; Reilly et al. 1997). The genetic loss is large and involves the gene-rich region around RB-1, D13S319, and D13S25 (Sinclair et al. 2001). It is possible that more than one gene is involved on chromosome 13 since Macdonald and colleagues (1999) reported a case of CIMF with a t(4;13)(q25;q12) and provided evidence for the involvement of a novel gene located at 13q12. The second and third most common abnormalities are deletions of 20q and partial duplication of the long arm of chromosome 1, respectively (Demory et al. 1988; Reilly et al. 1997). Amplifications of 1q follow a nonrandom pattern and, although it may involve the whole of 1q, it always appears to include the specific segment, 1q23-1q32 (Donti et al. 1990). The inability to identify common breakpoints, or a preferential translocation site, suggests that an increase in gene(s) copy number located on 1q is more important than the position effect due to the juxtaposition of specific DNA sequences. In support of this view, Zanke and colleagues (1994) have demonstrated amplification and overexpression of a hematopoietic protein tyrosine phosphatase (HePTP) in patients with partial trisomy 1q. The underlying molecular consequences of 13q- and 20q- remain to be determined, although extensive mapping and mutational screening have not identified any candidate genes and suggest that haplo-insufficiency may be a mechanism (reviewed Reilly 2005). These three lesions, however, are not specific for CIMF and have also been reported in polycythemia vera, myelodysplastic syndrome, and other hematological malignancies. In contrast, the abnormality der(6)t(1;6)(q23-25;p21-22) has been recently identified as a possible

marker for CIMF, although it is scarce, occurring in less than 3% of cases (Dingli et al. 2005). The incidence of chromosomal abnormalities in CIMF is significantly lower in younger patients (Cervantes et al. 1998), a fact that may explain their better prognosis. Indeed, normal cytogenetic findings are characteristic of pediatric cases, which, coupled with their long-term survival, suggests that they may have a different pathogenesis and require a more conservative management (Altura et al. 2000). Comparative genomic hybridization (CGH) studies have revealed that genomic aberrations are much more common than indicated by standard cytogenetic analysis and occur in the majority of cases. Gains of 9p appear to be the most frequent finding, occurring in 50% of cases, and suggests that genes on 9p may play a crucial role in the pathogenesis of CIMF (Al-Assar et al. 2005). A third of patients with CIMF possess an abnormal karyotype at diagnosis (Okamura et al. 2001; Reilly et al. 1994), although this increases to approximately 90% following acute transformation, a finding that supports the multistep process of leukemogenesis (Mesa et al. 2005; Reilly et al. 1994). The majority of leukemic transformations exhibit "high risk" cytogenetic changes, including -5/5q- and -7/-7q and, as a result, respond dismally to chemotherapy (Mesa et al. 2005).

Chromosomal abnormalities have been associated with a poor prognosis in several studies (Demory et al. 1988; Reilly et al. 1997), although the prognostic impact of specific cytogenetic lesions has been difficult to define. A recent report addressed this issue and indicated that only certain clonal abnormalities, such as trisomy 8 and deletion of 12p, carry an adverse prognosis, in contrast to the majority of changes which have little survival effect (Tefferi et al. 2001a). In addition, a number of rare karyotypic abnormalities, unrelated to therapy, have been associated with a poor outcome. Trisomy 13, a nonrandom aberration in myelofibrosis, confers a poor prognosis due to early blast transformation (Zojer et al. 1999), as appears also to be the case for del(1)t(1;9) (Rege-Cambrin et al. 1991) and t(6;10)(q27;q11) (Cox et al. 2001). Recently, Strasser-Weipel et al. (2004) reported the association of chromosome 7 deletions (-7/7q-) with an unfavorable prognosis, although surprisingly not with leukemic transformation. Finally, cytogenetic abnormalities have also been linked to treatment response, with anemia responding less well in patients with chromosomal abnormalities (Besa et al. 1982).

15.2.3 Molecular Studies

Recently, an acquired somatic point mutation in the *JAK2* gene (Val617Phe) has been reported in 49% of a total of 88 CIMF patients by four independent groups (Baxter et al. 2005; James et al. 2005; Kralovics et al. 2005; Levine et al. 2005). This mutation, which also occurs in approximately 90% of patients with polycythemia vera and 40% of patients with essential thrombocythemia, almost certainly contributes to the myeloproliferative state, as cellular expression has been shown to lead to growth factor independence (James et al. 2005) as well as myelofibrosis in a murine bone marrow transplant model (Wernig et al. 2006). Interestingly, 22% of CIMF cases are homozygous for the *JAK2* mutation, a feature that appears linked to loss of heterozygosity of 9p (Kralovics et al. 2005). Initial clinical studies suggest that CIMF patients possessing the *JAK2* mutation have a higher total white cell and neutrophil count, are less likely to require blood transfusions and have a poorer survival (Campbell et al. 2006). It is to be hoped that this novel finding will lead to the future development of targeted therapy for use in this group of related disorders. The molecular defects in the remaining cases remain essentially unknown. Intriguingly, *STAT5* has been reported to be constitutively activated in the majority of CIMF CD34⁺ cells and megakaryocytes (Komura et al. 2003), and suggests that *STAT5* activation may occur by mechanisms other than by acquired *JAK2* mutations. However, mutational screening of candidate receptor tyrosine kinase (RTK) genes that activate *JAK2*, namely *c-KIT*, *c-FMS*, and *FLT3*, has been unhelpful (Abu-Duhier et al. 2003). A possible clue to alternative *STAT5* activation mechanisms in CIMF is the reported overexpression of FK506 binding protein 56 (FKBP51) in megakaryocytes. This immunophilin is known to induce sustained activation of the *JAK2/STAT5* pathway as well as being able to induce an antiapoptotic phenotype (Giraudier et al. 2002). Overexpression of FKBP51 may also have a role in the activation of NF- κ B, a feature of CIMF megakaryocytes and circulating CD34 cells (Komura et al. 2005). The mechanism by which FKBP51 is upregulated in CIMF, however, remains to be determined. *RAS* mutations, predominantly affecting codon 12 of *N-RAS*, have been described, but appear rare, occurring in approximately 6% of patients in chronic phase (Reilly et al. 1994). Mutations involving *p53* and *p16* are also rare in the chronic phase of the disease, although they may be associated with transformation of a variety of BCR-ABL-

negative chronic myeloproliferative disorders, including myelofibrosis (Gaidano et al. 1993; Tsuruni et al. 2002; Wang and Chen 1999). Kimura and colleagues (1997) reported *KIT* mutations (Asp52Asn) in two patients and suggested that this acquired abnormality resulted in enhanced sensitivity to *KIT* ligand. However, a detailed study did not confirm these findings, suggesting that such mutations are rare (Abu-Duhier et al. 2003). Loss of heterozygosity (LOH) studies have highlighted *RAR β 2* to be a candidate tumor suppressor gene in CIMF, although for most patients epigenetic changes rather than gene deletion may be the most significant determinant of reduced activity (Jones et al. 2004). Finally, a recent study, using oligonucleotide microarrays on purified CD34⁺ cells, has highlighted the potential underlying complexity in CIMF by identifying 95 genes that were aberrantly expressed (Jones et al. 2005).

15.2.4 Role of Growth Factors

Myelofibrotic stroma has a complex structure, characterized by an increase in total collagen, that includes both the interstitial and basement membrane collagens, types I, III, IV, V, and VI (Apaja-Sarkkinen et al. 1986; Gay et al. 1984; Reilly, et al. 1985a, 1995b). In addition, there is an excessive deposition of fibronectin (Reilly et al. 1985a), laminin (Reilly et al. 1985b) tenascin (Reilly et al. 1995), and vitronectin (Reilly and Nash 1988) as well as a marked neo-vascularization and an associated endothelial cell proliferation (Mesa et al. 2000; Reilly et al. 1985b). Indeed, the hypervascularity and sinusoidal hyperplasia leads to a marked increase in bone marrow blood flow (Charbord 1986). The increased deposition of interstitial and basement membrane antigens is supported by the findings of raised serum markers for laminin and collagen types I, III, and IV, especially in patients with active disease (Hasselbalch et al. 1986; Reilly et al. 1995). These complex structural features and the wealth of stromal proteins are now believed to result from the abnormal release of growth factors, especially PDGF and TGF- β , from clonally involved megakaryocytes (Fig. 15.1).

15.2.4.1 Platelet-Derived Growth Factor

A number of observations support the concept that the megakaryocytic lineage plays a pivotal role in the pathogenesis of myelofibrotic stroma. Structural and maturation

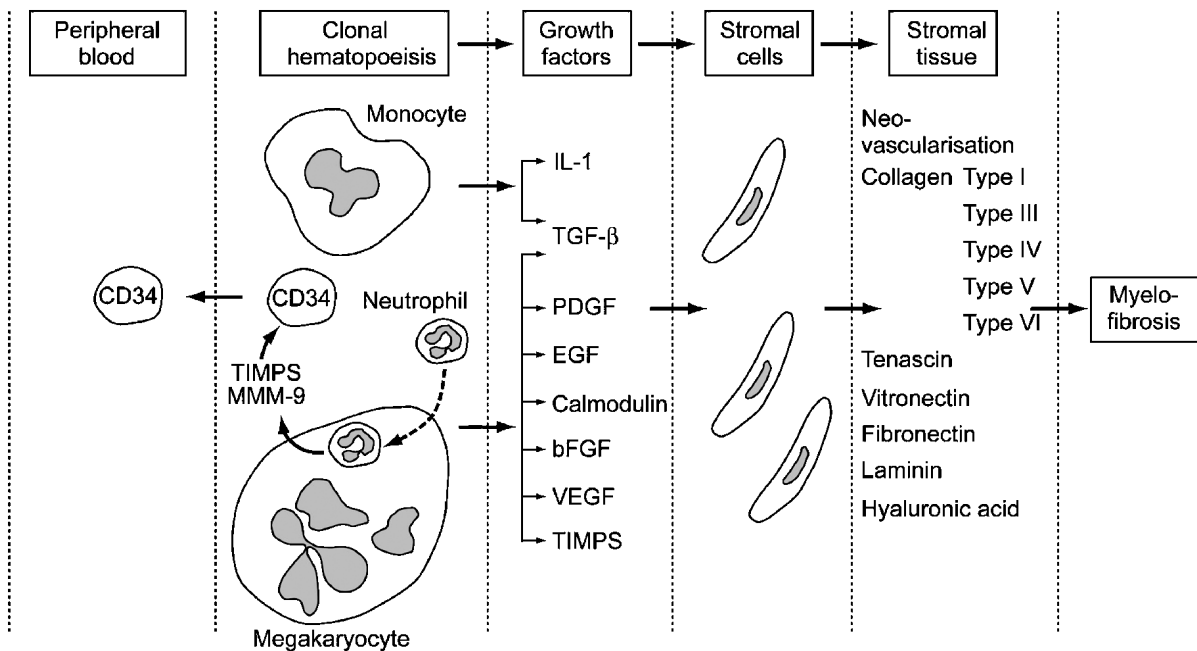


Fig. 15.1. The current pathogenetic model for the development of myelofibrotic stroma. bFGF, basic fibroblast growth factor; EGF, epidermal growth factor; IL-1, interleukin-1; PDGF, Platelet-derived

growth factor; TGF- β , transforming growth factor- β ; TIMPS, Tissue inhibitors of metalloproteinases; VEGF, vascular growth factor. (Modified from Reilly 1997, Blood Reviews 11:233–242)

tional defects of megakaryocytes are well-recognized features, including conspicuous proliferation and clustering, and with accumulation of fibrotic tissue often being associated with necrotic and/or dysplastic megakaryocytes (Thiele et al. 1991). In addition, bone marrow fibrosis is a well-described feature of patients with megakaryocytic leukemia (Den Ottolander et al. 1979) and the rare Gray Platelet Syndrome (Jantunen et al. 1994), disorders that are thought to affect platelet alpha granule packaging. However, the first tangible evidence for the role of megakaryocytic-derived growth factors was provided by Castro-Malaspina and colleagues (1981), who demonstrated that megakaryocytic homogenates stimulated the proliferation of bone marrow fibroblasts and that this effect was the result of PDGF. Subsequently, decreased platelet PDGF levels (Bernabei et al. 1986; Dolan et al. 1991; Katoh et al. 1988) associated with increased plasma and urinary levels (Gersuk et al. 1989) were reported in patients, a finding thought to reflect an abnormal release and/or leakage of PDGF from bone marrow megakaryocytes. In addition, similar findings for platelet β -thromboglobulin and platelet factor 4 favor a platelet and/or megakaryocyte release mechanism (Romano et al. 1990; Sacchi et al. 1986).

However, the release of PDGF, while undoubtedly inducing fibroblast growth, cannot account totally for the observed complexity of the stromal tissue. PDGF, for example, does not have angiogenic properties, nor does it increase the transcription of stromal proteins. Additional growth factors must play a role, the most important of which is probably transforming growth factor- β .

15.2.4.2 Transforming Growth Factor- β (TGF- β)

Like PDGF, TGF- β is synthesized by megakaryocytes, stored in platelet alpha granules and released at sites of injury (Fava et al. 1990). The pathological relevance of TGF- β lies in its ability to regulate extracellular matrix synthesis. It increases, for example, transcription of genes that code for fibronectin, collagens I, III and IV, and tenascin. It possesses powerful angiogenetic properties, with neovascularization occurring within 48 h of injection and, in addition, it can decrease the activity of metalloproteinases, enzymes that degrade extracellular stromal tissue (Overall et al. 1989; Roberts et al. 1986). In addition, TGF- β promotes endothelial cell migration, enhances stromal cell synthesis of vascular en-

dothelial growth factor (VEGF), and may also inhibit the production of antiangiogenic molecules (Harmey et al. 1998; O'Mahoney et al. 1998). The combined effect of these activities is the increased synthesis and accumulation of extracellular matrix. Evidence for a pathogenetic role in CIMF include the report of significantly increased intraplatelet TGF- β levels when compared to normal platelets (Martyr et al. 1991), the finding of active TGF- β synthesis by megakaryoblasts (Terui et al. 1990), and the finding of increased plasma concentrations in a case of acute micromegakaryocytic leukemia that correlated with enhanced stromal turnover (Reilly et al. 1993). In addition, TGF- β expression is increased in patients' peripheral blood mononuclear cells at the mRNA level and/or at the secreted protein level (Martyr et al. 1994). Megakaryocytes, however, may not be the only cellular source of TGF- β , since TGF- β deposition appears to correlate with fibrosis even in cases with normal or reduced megakaryocyte numbers (Johnson et al. 1995). Interestingly, macrophages are frequently increased in myelofibrosis (Thiele et al. 1992; Titius et al. 1994) as is serum M-CSF, a growth factor which regulates the survival, proliferation, differentiation, and activation of macrophages (Gilbert et al. 1989). Furthermore, it has been shown that circulating monocytes in CIMF may be preactivated and contain increased levels of cytoplasmic TGF- β and IL-1 (Rameshwar et al. 1994). It has also been hypothesized that extracellular matrix protein-adhesion molecule interactions, involving CD44, may induce overproduction of fibrogenic cytokines in CIMF monocytes and contribute to stromal fibrosis in the bone marrow (Rameshwar et al. 1996). However, TGF- β is known to negatively regulate the cycling status of primitive progenitor cells and yet CIMF is characterized by an increased number of circulating CD34+ cells. This apparent paradox has been addressed by Le Bousse-Kerdiles and colleagues, who suggest that the explanation may, in part, be due to an acquired reduction in TGF- β type II receptor expression on myelofibrotic CD34+ progenitor cells. This fact, coupled with increased expression of basic fibroblast growth factor (bFGF) on the same cells, could explain the impaired inhibition by TGF- β (Le Bousse-Kerdiles et al. 1996).

15.2.4.3 Additional Growth Factors and Cytokines

A number of additional growth factors have been implicated in the pathogenesis of myelofibrotic stroma, including the calcium binding protein calmodulin, basic fibroblast growth factor (bFGF), vascular endothelial growth factor (VEGF), and the tissue inhibitors of metalloproteinases (TIMPS) (Fig. 15.1). Several facts suggest a role for extracellular calmodulin, including the finding of elevated urinary levels in CIMF, the knowledge that platelets are a rich source of calmodulin, and the fact that the protein acts as a fibroblast mitogen in the absence other growth factors (Dalley et al. 1996; Eastham et al. 1994). The finding of elevated plasma levels of VEGF in CIMF (Novetsky et al. 1997), coupled with the fact that megakaryocytes produce and secrete large amounts (Brogi et al. 1994), suggests that this multifunctional cytokine may also contribute to the pathogenesis of the characteristic neoangiogenesis. In addition, bFGF has been reported by Martyr and colleagues (1997) to be elevated in platelets and megakaryocytes from CIMF patients, while urinary excretion is similarly increased (Dalley et al. 1996). Megakaryocytes and platelets are also rich sources of releasable TIMPS, with serum levels being significantly higher than those found in plasma. These proteins may contribute to the induction of marrow fibrosis by inhibiting connective tissue breakdown by members of the matrix metalloproteinase family and by functioning as growth factors for marrow fibroblasts. Indeed, Murate and colleagues (1997) have shown that the combined effects of TIMP-1 and TIMP-2 are almost equal to the fibrogenic effects of TGF- β . Recently, Emadi and colleagues (2005) have provided evidence for the involvement of IL-8 and its receptors (CXCR1 and CXCR2) in the altered megakaryocytic proliferation, differentiation and ploidyization characteristic of CIMF, while IL-6 is also likely to be involved (Wang et al. 1997). The study of the pathogenesis of osteosclerosis typical of advanced CIMF has been limited, but it may be related to the overproduction of osteoprotegerin (OPG) (Wang et al. 2004).

Finally, an underlying mechanism for megakaryocyte-derived growth factor release has recently been proposed, in addition to the standard model of dysplasia and defective alpha granule packaging. CIMF, for example, is characterized by enhanced neutrophil and eosinophil emperipolesis by megakaryocytes, with the latter expressing both abnormal amounts and distribution

of P-selectin, an important mediator of neutrophil rolling (Schmitt et al. 2002). Activation of the engulfed neutrophils results in release of their proteolytic enzymes leading both to death of cells and the release of megakaryocytic TGF- β and PDGF. This phenomenon could also underlie the increased neutrophil elastase and active MMP-9 present in CIMF which, as a result of their multiple proteolytic activities, may enhance the release of CD34⁺ progenitor cells from the bone marrow (Schmitt et al. 2002; Xu et al. 2003) (Fig. 15.1).

15.2.5 Animal Models

Several mouse models support the pivotal role of megakaryocytes in the development of the stromal proliferation, or myelofibrosis, that characterizes CIMF. These models were originally developed to investigate the role of thrombopoietin (TPO) and its receptor (Mpl), as well as the transcription factor GATA-1, in the control of megakaryocytopoiesis (Vannuchi et al. 2004; Yan et al. 1996). It was noted, however, that mice that overexpressed TPO, or underexpressed GATA-1, developed a clinical state similar to myelofibrosis, with tear drop poikilocytosis, increased circulating progenitors, and extramedullary hematopoiesis. The linking event appears to be a block in megakaryocyte differentiation, associated with an abnormal localization of P-selectin, which leads to neutrophil emperipoiesis and the eventual release of TGF- β 1 from megakaryocytic alpha granules (Vannucchi et al. 2005). These animal models support clinical observations and imply that myelofibrosis may not have a single cause, but may be the consequence of any perturbation that leads to increased neutrophil emperipoiesis within the megakaryocyte. Although such models do not provide any insight into the pathogenesis of the underlying clonal hematopoiesis, they do support the link between TGF- β and stromal tissue development and may be of value for identifying novel antifibrotic agents for use in reversing clinical myelofibrosis.

15.3 Diagnosis

Classical CIMF is characterized by bone marrow fibrosis, extramedullary hematopoiesis, splenomegaly, and a leuko-erythroblastic blood picture. However, in contrast to CML, there is no specific biological marker and many

Table 15.1. Conditions associated with bone marrow fibrosis

Malignant	Non-malignant
Chronic idiopathic myelofibrosis	Infections (e.g., TB, visceral leishmaniasis)
Other chronic myeloproliferative disorders, histoplasmosis, HIV) (e.g., PV, CML, ET)	Renal osteodystrophy
Acute megakaryoblastic leukemia	Vitamin D deficiency
(Acute myelofibrosis)	Hypothyroidism
Myelodysplastic syndromes	Hyperthyroidism
Acute myeloid leukemia	Gray platelet syndrome
Acute lymphoblastic leukemia	Systemic lupus erythematosus
Hairy cell leukemia	Scleroderma
Hodgkin's disease	Radiation exposure
Non-Hodgkin's lymphoma	Benzene exposure
Multiple myeloma	Gaucher's disease
Systemic mastocytosis	Osteopetrosis
Metastatic carcinoma (e.g., breast, prostate, stomach)	

Table 15.2. Italian Consensus Diagnostic criteria

Necessary criteria
Diffuse bone marrow fibrosis
Absence of Ph-chromosome or <i>BCR-ABL</i>
Optional criteria
Splenomegaly of any grade
Aniso-poikilocytosis
Presence of immature circulating myeloid cells
Presence of circulating erythroblasts
Clusters of megakaryocytes and abnormal megakaryocytes in the bone marrow
Myeloid metaplasia

Diagnosis of CIMF is acceptable if the following combinations are present: the two necessary criteria plus any other two optional criteria when splenomegaly is present, or the two necessary criteria plus any other four criteria if splenomegaly is absent.

studies have included a heterogeneous population of patients. The inappropriate inclusion of cases with secondary myelofibrosis, postpolycythemic myelofibrosis, and myelodysplasia with myelofibrosis and related disorders (Table 15.1) may explain the discrepancies in the early literature relating to cytogenetic abnormalities, therapeutic response, and prognosis. To address these difficulties, the “Italian Consensus Conference on Diagnostic Criteria for Myelofibrosis with Myeloid Metaplasia” proposed a definition of CIMF that has >80% sensitivity and specificity (Barosi et al. 1999). The definition requires two necessary criteria, namely diffuse bone marrow fibrosis and the absence of the Ph chromosome or *BCR-ABL* rearrangement, as well as a num-

ber of optional criteria (see Table 15.2). However, although this definition of CIMF encompasses a wide spectrum of the disease, from the early stages with slight reticulin fibrosis to the late osteomyelosclerotic phase (CIMF-1 to 3), it fails to include the recently recognized initial prefibrotic stage (CIMF-o) (see Figs. 15.2–15.4).

The concept of a prefibrotic stage of classical CIMF, that is distinguishable from essential thrombocythemia, has been stressed by a number of European histopathologists (Buhr et al. 2003; Georgii et al. 1998; Thiele and Kvasnicka 2004; Thiele et al. 1999) with the result that prefibrotic CIMF has been incorporated in the WHO classification of hematopoietic and lymphoid tumors

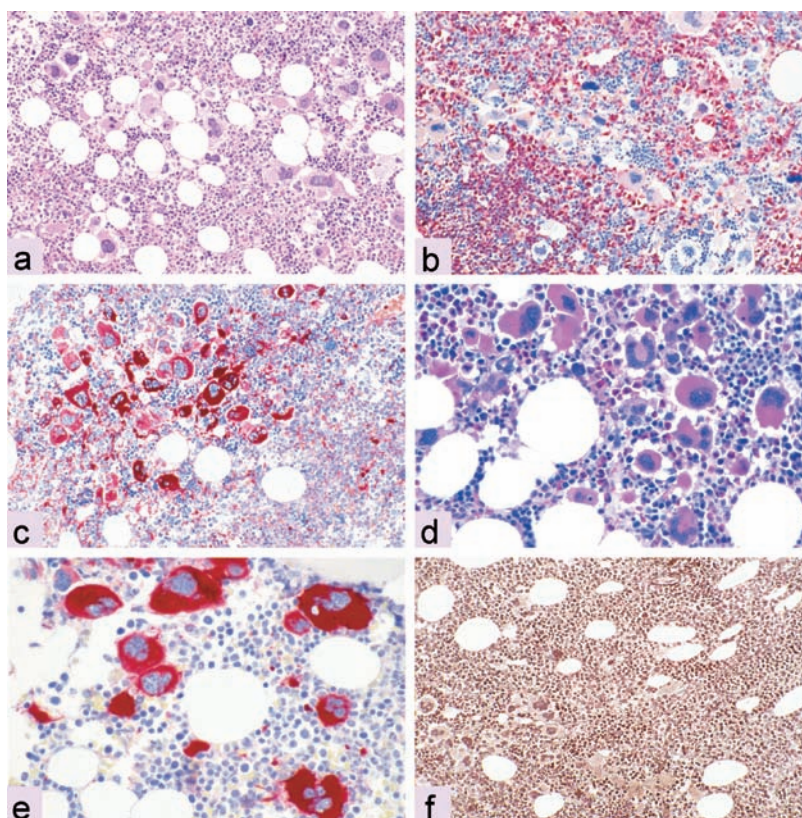


Fig. 15.2. Prefibrotic CIMF (CIMF-0). (a) An overall hypercellularity is evident including prominent growth of abnormally differentiated megakaryocytes (i.e., false ET). (b) There is a mixed neutrophil granulocytic and megakaryocytic proliferation with loose to dense clustering. (c) Atypias of megakaryopoiesis include histotopography (dense clustering) besides maturation defects revealing hypobulbated (bulbous) and hyperchromatic nuclei. (d) A prevalence of abnormal megakaryocytes with deviation of nuclear-cytoplasmic ma-

ture is detectable. (e) Megakaryocytic abnormalities are highlighted by application of immunohistochemistry. (f) No increase in the reticulin fiber content may be observed. (a, b, c, f) $\times 70$; (d, e) $\times 380$; (a), hematoxylin-eosin, (b), AS-D-chloroacetate esterase, (c) and (e), CD61 immunostaining, (d), PAS (periodic acid Schiff reagent), (d), Silver immunostaining after Gomori (courtesy of Dr. Kvasnicka)

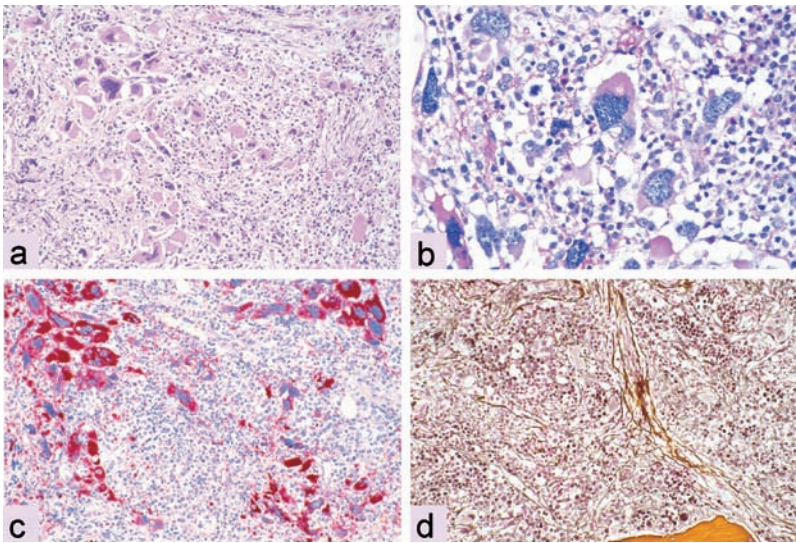


Fig. 15.3. Manifest CIMF. (a) In addition to a still slightly hypercellular bone marrow and a prominent granulopoiesis there are clusters of atypical megakaryocytes trapped in a fibrous meshwork. (b) Megakaryocytes show abnormal cloud-like (bulbous) nuclei and maturation defects (c) Dense clustering of atypical megakaryocytes is a

conspicuous feature. (d) A dense increase in reticulin and some collagen fibers are characteristic. (a, c, d) $\times 170$; (b) $\times 380$; (a) Hematoxylin-eosin, (b) PAS. (c) CD61 immunostaining (d) Silver impregnation after Gomori (courtesy of Dr. Kvasnicka)

(Thiele et al. 2001b). It has been estimated that approximately 25% of patients with CIMF initially present with a hypercellular bone marrow characterized by granulocytic and megakaryocytic proliferation and with little or no reticulin. The diagnosis requires careful examination of the bone marrow trephine and relies on the identification of morphologically atypical megakaryocytes, including dense clustering with hypolobulated (bulbous) and hyperchromatic nuclei. Other diagnostically important parameters include the frequency and shape of microvasculature (Kvasnicka et al. 2004), the level of $CD34^+$ progenitor cells (Thiele and Kvasnicka 2002), and abnormalities of cell kinetics (Kvasnicka et al. 1999). Prefibrotic CIMF is likely to have been misdiagnosed as essential thrombocythemia in many studies since it is characterized by thrombocythemia, borderline anemia, mild splenomegaly, and an absence of a leuko-erythroblastic blood picture (Thiele and Kvasnicka 2004; Thiele et al. 2001a). The natural history of prefibrotic CIMF remains unclear since prospective studies are lacking. However, preliminary data suggest that the rate of progression to advanced disease may depend on the degree of megakaryocytic dysplasia (Buhr et al. 2003; Thiele et al. 2003).

15.4 Clinical Manifestations

CIMF characteristically occurs after the age of 50, with a median age at diagnosis of approximately 60 years. About 25% of patients are asymptomatic at diagnosis and are identified following routine examination. The most common symptoms in classical CIMF are the consequence of anemia, namely fatigue, weakness, dyspnea, and palpitations. Splenomegaly is characteristic and when massive can lead to a variety of complaints including abdominal discomfort and early satiety (Fig. 15.5). Splenic infarction, due to the inability of the blood supply to match organ growth, usually produces transient discomfort although rarely can result in severe abdominal pain simulating an abdominal emergency (Fig. 15.6). Hepatomegaly occurs in approximately 70% of cases and portal hypertension may result from increased hepatic blood flow or intrahepatic obstruction (Tsao et al. 1989; Wanless et al. 1990). Nonspecific symptoms may dominate the clinical picture in CIMF, including low-grade fever, night sweats, and weight loss, and are associated with a poor prognosis (Cervantes et al. 1998). Patients may also complain of bone pain, especially in the lower extremities. Bleeding may complicate the clinical course and although often mild, manifesting

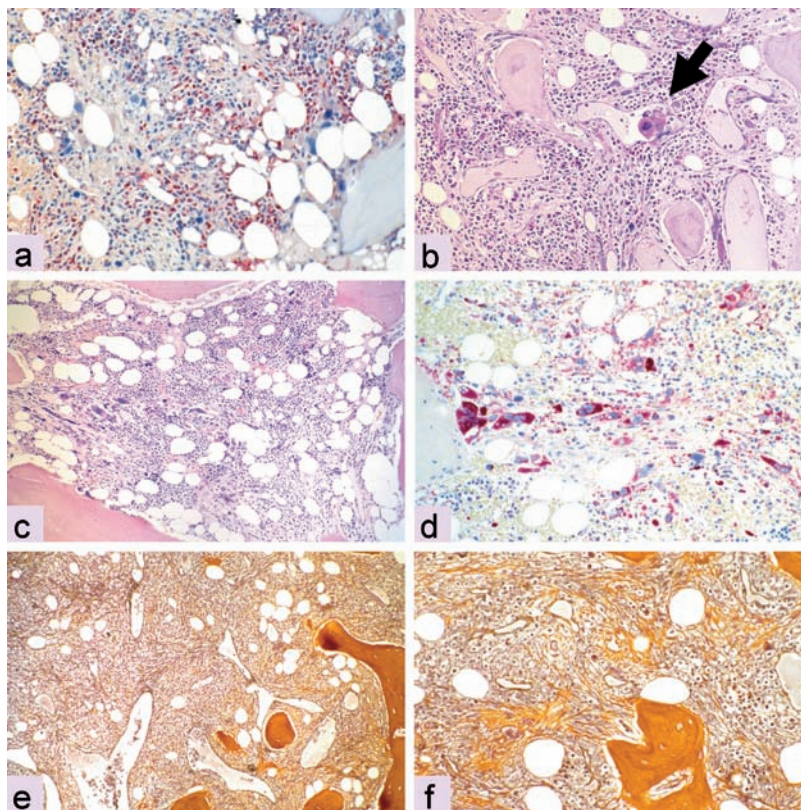


Fig. 15.4. Advanced CIMF. (a) Patchy hematopoiesis shows megakaryocyte clusters besides a reduction of granulo- and erythropoiesis and bundles of fibers. (b) Prominent dilated sinuses with intraluminal megakaryopoiesis (arrow) may be observed. (c) In addition to differences in cellularity there are initial plaque-like osteosclerotic changes and a meshwork of fibers. (d) Megakaryo-

cytes reveal abnormalities of histopography (endosteal translocation and clustering) apparently in close association with bud-like endophytic bone formation – osteosclerosis may usually be found. (a, b, d) $\times 170$; (c, e, f) $\times 80$; (a) AS-D-chloroacetate esterase, (b) PAS, (c) Haematoxylin-eosin, (d) CD61 immunostaining; (e, f) Silver impregnation after Gomori (courtesy of Dr. Kvasnicka)

as petechiae and ecchymoses, can be life threatening due to massive gastrointestinal hemorrhage. The hemorrhagic diathesis may result from a combination of thrombocytopenia, acquired platelet dysfunction, and low-grade disseminated intravascular coagulation.

Extramedullary hematopoiesis (EMH), or myeloid metaplasia, may result in a bewildering array of symptoms which depend on the specific organ involved. EMH, for example, may affect the central nervous system and result in spinal cord compression (Horwood et al. 2003; Price and Bell 1985), delirium (Cornfield et al. 1983), diabetes insipidus (Badon et al. 1985), serious headaches and exophthalmos due to meningeal infiltration (Ayyildiz et al. 2004; Landolfi et al. 1988), as well as raised intracranial hypertension with papilledema and ultimately coma (Cameron et al. 1981; Ligumski et al.

1979; Lundh et al. 1982). Involvement of lymph nodes can lead to generalized and marked lymphadenopathy (Williams et al. 1985). Pleural infiltration may result in hemothoraces (Kupferschmid et al. 1993) and pleural effusions (Jowitt et al. 1997) (Fig. 15.7), while massive ascites may result from ectopic implants of peritoneal or mesenteric extramedullary hematopoiesis (Yotsumoto et al. 2003). The effusions often contain a variety of hematopoietic elements, including megakaryocytes, immature myeloid cells, and erythroblasts. The gastrointestinal tract may be involved and this results in abdominal pain and intestinal obstruction (Mackinnon et al. 1986; Sharma et al. 1986), while infiltration of the kidneys (Fig. 15.8), prostate, and gallbladder have been reported to result in chronic renal failure, bladder outlet obstruction, and chronic cholecystitis, respectively

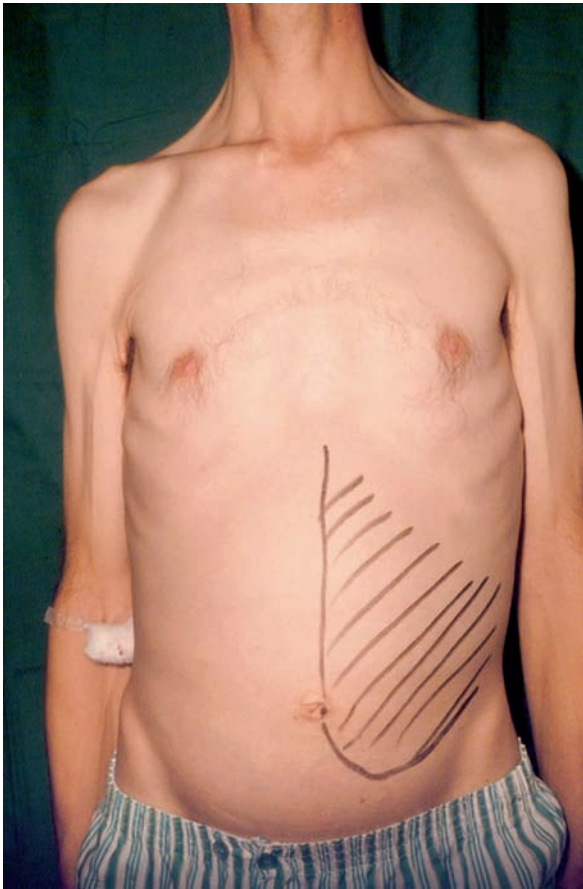


Fig. 15.5. Gross splenomegaly (extending 22 cm below the left costal margin) and associated cachexia

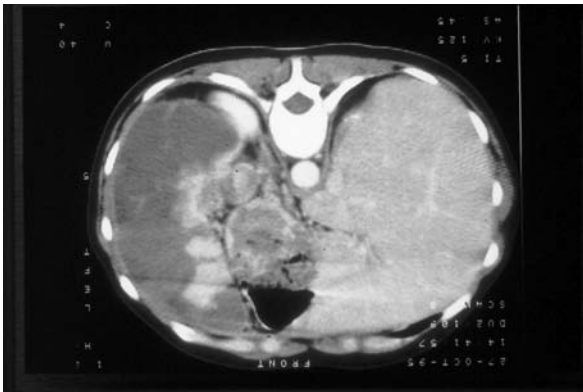


Fig. 15.6. Massive splenic infarction necessitating splenectomy

(Humphrey and Vollmer 1991; Schnuelle et al. 1999; Thorns et al. 2002). Involvement of breast tissue may mimic carcinoma (Martinelli et al. 1983), while urethral

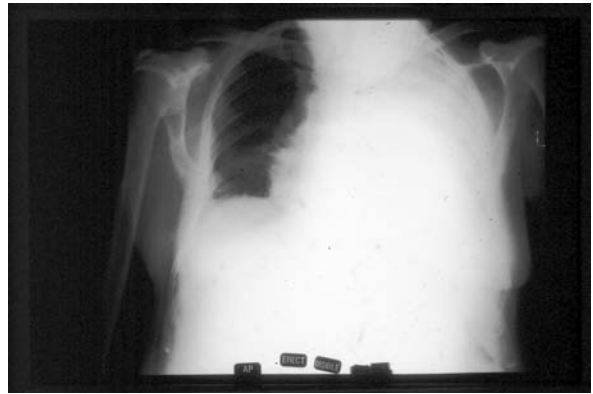


Fig. 15.7. Pleural effusion in a patient with myelofibrosis and extramedullary hematopoiesis involving the pleura

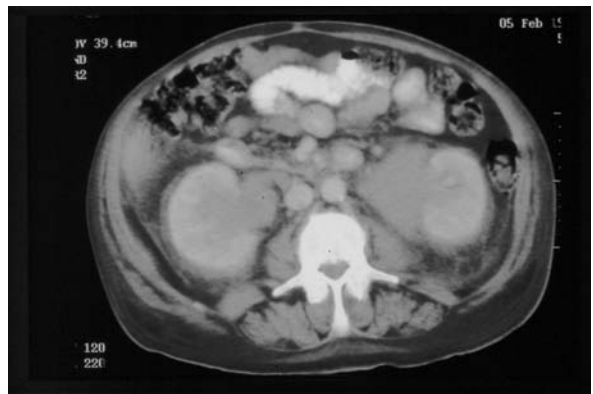


Fig. 15.8. Extramedullary hematopoiesis involving both kidneys

infiltration has been reported to masquerade as a caruncle (Balogh and O'Hara 1986) and synovial involvement can give rise to arthritis (Heinicke et al. 1983). Skin manifestations are rare and include erythematous plaques (Fig. 15.9), nodules, diffuse or papular erythema, ulcers, and bullae (Loewy et al. 1994). Rarely, the development of CIMF can be preceded by the presence of neutrophilic dermatosis, or "Sweet's syndrome" while pyoderma gangrenosum and leukemic infiltrations have been reported (Gibson et al. 1985).

The major causes of death are infection, hemorrhage, cardiac failure, and acute leukemic transformation. The latter, which occurs in approximately 15% of patients (Silverstein et al. 1973) are commonly myeloblastic or myelomonoblastic, but may involve the megakaryocytic (Reilly et al. 1993), erythroid (Garcia et al. 1989), lymphoid (Polliak et al. 1980), and basophilic



Fig. 15.9. Cutaneous extramedullary hematopoiesis

lineages (Sugimoto et al. 2004). Hernandez and colleagues (1992) have described the occurrence of mixed myeloid (myeloblastic-erythroid-megakaryocytic) or hybrid (myeloid-lymphoid) phenotypes in up to a third of cases, a fact that reflects the pluripotent stem cell origin of the disease. Localized granulocytic sarcomas, or chloromas, can develop in a wide variety of sites, including bone, lymph nodes, and skin and on occasion precede the diagnosis of leukemia.

15.5 Laboratory Features

A normocytic normochromic anemia is characteristic of classic CIMF, as is anisocytosis, poikilocytosis, and teardrop-shaped cells (dacrocytes). The origin of dacrocytes is uncertain but they are thought to be the sine qua non of extramedullary hematopoiesis. Nucleated red cells are present in the peripheral blood of nearly all cases and there is frequently a mild reticulocytosis. The major cause of anemia is ineffective erythropoiesis but other causes may include iron deficiency, red cell sequestration, and hemodilution due to plasma volume expansion as a result of splenomegaly. Hemolysis can be significant and, although frequently direct antiglobulin test (DAT) negative, may be autoimmune in etiology (Bird et al. 1985).

Immunological abnormalities, other than anti-red cell antibodies, are common and include the development of antinuclear and rheumatoid antibodies, lupus-type anticoagulants, antiphospholipid antibodies (Rondeau et al. 1983), hypocomplementemia (Gordon et al. 1981), and increased nodules of lymphoid cells in the bone marrow (Caligaris Cappio et al. 1981). Lewis and

Pegrum (1972) described immune complexes on leukocytes while, more recently, antibodies directed against stromal proteins, for example anti-Gal antibodies, have been demonstrated that appear to correlate with disease activity. Interestingly, galactosidic determinants are thought to be expressed by fibroblasts and megakaryocytes in patients with CIMF raising the possibility of an autoimmune pathogenesis (Leoni et al. 1993). A number of these abnormalities are likely to be epiphenomena, resulting from impaired reticulo-endothelial system clearance, but immunological mechanisms have been postulated for the induction and/or maintenance of the disease (Caligaris Cappio et al. 1981); for example, immune complexes could result in platelet activation and additional growth factor release. Interestingly, Gordon and colleagues (1981) noted a correlation between circulating immune complexes and disease activity as manifested by increased transfusion requirements, bone pain, and fever. The immunological hypothesis for myelofibrosis is supported by reports of successful immunosuppressive therapy, including low-dose dexamethasone (Jack et al. 1994), prednisolone (Mesa et al. 2003) and cyclosporin A (Pietrasanta et al. 1994), as well as the reports of myelofibrosis associated with systemic lupus erythematosus (Kaelin and Spivak 1986) and polyarteritis nodosa (Connelly et al. 1982).

15.6 Prognosis

The overall median survival of classical CIMF varies from series to series but is approximately 4 years (Demory et al. 1988; Reilly et al. 1997; Rupoli et al. 1994; Varki et al. 1983), although individual survival may range from 1 to over 30 years. This is considerably lower, however, than the 14-year median survival of age- and sex-matched controls (Rozman et al. 1991). As a result, many groups have used univariate or multivariate analysis to identify clinical and laboratory features that predict survival. Despite the bewildering number of prognostic factors highlighted, most studies agree on the predictive value of anemia (Barosi et al. 1988; Cervantes et al. 1991; Dupriez et al. 1996; Ivanyi et al. 1984; Kreft et al. 2003; Njoku et al. 1983; Reilly et al. 1997; Rupoli et al. 1994; Visani et al. 1990), age at diagnosis (Barosi et al. 1988; Cervantes et al. 1997; Kvasnicka et al. 1977; Reilly et al. 1997; Varki et al. 1983), karyotype (Dupriez et al. 1996; Reilly et al. 1997; Tefferi et al. 2001), and the percentage of immature granulocytes and/or circulating myelo-

blasts (Barosi et al. 1988; Cervantes et al. 1991; Visani et al. 1990). The data regarding the prognostic value of absolute peripheral blood CD34+ counts, however, are less clear. Several groups, for example, have suggested that, in addition to a possible diagnostic role, elevated CD34+ counts are an important adverse prognostic marker (Barosi et al. 2001; Passamonti et al. 2003; Sagaster et al. 2003). In contrast, Arora and colleagues (2004), although finding that counts above $0.1 \times 10^9/L$ correlated with shortened survival, noted that this significance was lost on multivariate analysis. Finally, the degree of angiogenesis (Mesa et al. 2000), in contrast to the extent of collagen fibrosis or osteosclerosis (Dupriez et al. 1996; Kvasnicka et al. 1997; Rupoli et al. 1994), has been shown to be a significant and independent risk factor for overall survival.

Two simple and practicable schemas have been reported that allow the identification of patients with limited life expectancy, for whom more aggressive therapeutic approaches might be appropriate (Dupriez et al. 1996; Reilly et al. 1997). The most widely used is the Lille scoring system (Table 15.3) which is based on two adverse prognostic factors, namely hemoglobin $<10g/dL$ and a total white count <4 or $>30 \times 10^9/L$, and which separates patients into three groups with low (0 factor), intermediate (1 factor), and high risk (2 factors) disease, associated with median survivals of 93, 26, and 13 months, respectively (Dupriez et al. 1996). The Sheffield schema (Table 15.4), by combining age, hemoglobin concentration, and karyotype, identifies patient groups with median survival times that vary from 180 months (good risk) to 16 months (poor risk) (Reilly et al. 1997). However, there are two important caveats that apply to these and many other studies. Firstly, only a few groups have included the full spectrum of the

Table 15.4. The SHEFFIELD schema for predicting survival (reproduced from Reilly et al. 1997 with permission)

Age (years)	Hb (g/dl)	Karyotype	Median survival (months) (95% CI)
<68	<10	N	54 (46, 62)
		A	22 (14, 30)
	>10	N	180 (6, 354)
		A	72 (32, 112)
>68	<10	N	44 (31, 57)
		A	16 (5, 27)
	>10	N	70 (61, 79)
		A	78 (26, 130)

Demonstrating median survival times in months with associated 95% confidence intervals in parenthesis;

N, normal; A, abnormal.

disease, from the early prefibrotic phase to the advanced full-blown osteomyelosclerotic state. This is important as the early prefibrotic stages of CIMF show a more favorable outcome than the advanced stages of disease (Kvasnicka et al. 1997). Secondly, most studies have included very few young patients, a fact that could potentially limit the schema's utility when attempting to identify cases suitable for bone marrow transplantation. This deficiency has been addressed by Cervantes and colleagues (1999), who reported a large collaborative study of 116 patients below the age of 55 years and concluded that, by using a combination of hemoglobin, constitutional symptoms, and percentage of blasts, patients with low- and high-risk disease could be identified. Importantly, the median survival in this cohort was 128 months which is significantly better than that reported for studies of unselected patients.

Table 15.3. The LILLE scoring system

No. of adverse prognostic factors	Risk group	Cases (%)	Median survival (months)
0	Low	47	93
1	Intermediate	45	26
2	High	8	13

Adverse prognostic factors; Hb $<10g/dL$, WBC <4 or $>30 \times 10^9/L$. (Reproduced from Dupriez et al. 1997 with permission).

15.7 Management

15.7.1 Medical Therapy

15.7.1.1 Cytotoxic Therapy

Cytotoxic chemotherapy has a definite role in the management of CIMF patients. Hydroxyurea, the most widely used agent (Lofvenberg et al. 1990; Manoharan 1991), can reduce the degree of hepatosplenomegaly, decrease or eliminate constitutional symptoms, reduce thrombocytosis and, in some cases, lead to an increase in hemoglobin. Hydroxyurea may also be useful in individuals who develop compensatory hepatic myeloid metaplasia following splenectomy and it has also been shown to improve bone marrow fibrosis (Lofvenberg et al. 1990). The use of busulfan has been reported in the proliferative phase of the disease (Manoharan and Pitney 1984), but the risks of prolonged cytopenias are significant. Responses are often short-lived, lasting a median of only 4.5 months (Silverstein 1975). Low-dose melphalan (starting at 2.5 mg three times a week) may be an alternative option (Petti et al. 2002) but again hematological toxicity is common. 2-chlorodeoxyadenosine (2-CdA) may have a palliative role in controlling the extreme thrombocytosis and leukocytosis, as well as the accelerated hepatomegaly that can occur post splenectomy. Responses were observed in about half of patients and occurred in most cases by the second course (Faoro et al. 2005; Tefferi et al. 1997).

15.7.1.2 Androgens

Anemia, usually normochromic normocytic, is a common problem in CIMF, with 20–25% of presenting cases being symptomatic. Iron deficiency, ineffective hematopoiesis, erythrocytic sequestration, hemodilution secondary to plasma volume expansion, and hemolysis are recognized mechanisms. Patients with normal red cell masses and marked increase in plasma volume have a dilutional form of anemia that does not require treatment. Androgen therapy, including nandrolone, fluoxymesterolone, and oxymetholone, improves marrow function in approximately 40% of patients (Besa et al. 1982; Brubaker et al. 1982; Hast et al. 1978), with optimal responses seen in patients lacking massive splenomegaly and in those with a normal karyotype (Besa et al. 1982). Danazol (400–600 mg/day), a synthetic attenuated androgen, may give similar results with the added benefit of correcting thrombocytopenia and reducing

the degree of splenomegaly in some patients (Cervantes et al. 2000, 2005; Levy et al. 1996). Androgen therapy should be continued for a minimum of 6 months and once a response is obtained, it should be reduced to the lowest maintenance dose. Pretreatment variables associated with response to danazol include lack of transfusion requirement and higher hemoglobin concentration at commencement of treatment (Cervantes et al. 2005). Side effects include fluid retention, increased libido, hirsutism, abnormal liver function tests, and hepatic tumors. All treated patients should have regular monitoring of liver function tests and periodic abdominal ultrasound investigation to detect liver tumors. In addition, male patients should be screened for prostate cancer prior to therapy.

15.7.1.3 Erythropoietin

Human recombinant erythropoietin (EPO) has been shown by several groups to be an effective and safe therapy in CIMF, although the number of reported cases remains small (Aloe-Spiriti et al. 1993; Bourantas et al. 1996; Hasselbalch et al. 2002; Tefferi and Silverstein 1994). Hasselbalch et al. (2002), for example, reported that 90% (9 of 10 evaluable cases) attained a favorable response, which was maintained in the majority of patients. Importantly, most responding individuals exhibited inappropriately low serum EPO levels for the degree of anemia. More recently, Cervantes et al. (2004) confirmed these findings and showed that 45% of patients responded favorably to a dose of 10,000 U three times a week. In addition, those with a serum EPO level <125 U/L and those that were transfusion independent had a more favorable outcome. It can be concluded from these studies that EPO is a well-tolerated therapy for the anemia in CIMF but that its use should be restricted to cases with inappropriately low serum EPO levels. It should be noted, however, that the majority of patients have appropriate levels for the degree of anemia (Barosi et al. 1993a). The dose may be doubled if there has been no response after 1–2 months and the treatment discontinued if there has been no response after 3–4 months.

15.7.1.4 Interferon

Parmeggiani et al. (1987) reported the use of α -interferon (α -IFN) for the treatment of painful splenomegaly in CIMF. Splenic pain and pressure symptoms resolved with a decrease in spleen size, although peripheral

counts deteriorated. Pegylated IFN, a polyethylene glycol formulation of α -IFN that is administered once a week, is currently undergoing clinical trials in CIMF and may have the advantage of being better tolerated (Verstovsek et al. 2003).

15.7.1.5 Thalidomide

Recently, thalidomide has been advocated as a therapy for controlling angiogenesis in several neoplastic and inflammatory diseases. The marked neo-vascularization that characterizes CIMF bone marrow (Mesa et al. 2000, Reilly et al. 1985b) suggested that thalidomide might be beneficial in this disease and has led to several small studies. A pooled analysis of the latter indicated that thalidomide can ameliorate anemia, thrombocytopenia, and splenomegaly in some cases, but that most patients were intolerant of standard doses (200–800 mg/day), with nearly 50% of cases withdrawing from the studies by the third month (Barosi et al. 2002). As a result, the combination of low-dose thalidomide (50 mg/day) and prednisolone at 0.5 mg/kg/day slowly tapered over the course of 3 months was evaluated and shown to be associated with a higher response rate and lower toxicity (Mesa et al. 2003). A meaningful improvement in anemia was demonstrated in 62% of all patients, while a 70% response rate was obtained for those cases that were transfusion dependent. However, although thrombocytopenia and splenomegaly improved in 75% and 19% of cases, respectively, there was no apparent decrease in extramedullary hematopoiesis or angiogenesis, suggesting that thalidomide's activity may not be due to its antiangiogenic properties. Following the discontinuation of prednisolone, the improvement in anemia and thrombocytopenia was lost in over a third of cases. The role of steroids in this study supports earlier data that indicated the benefit of low-dose dexamethasone (Jack et al. 1994). A recent European study (Marchetti et al. 2004) confirmed the usefulness of single agent low-dose thalidomide (50 mg/day) in a cohort of 63 patients with advanced stage disease and concluded that it was effective especially for transfusion-dependent and/or thrombocytopenic patients and for those requiring control of progressive splenomegaly. A combination of thalidomide and erythropoietin has been shown to correct the anemia in some cases in which both drugs have previously failed as single agents (Visani et al. 2003). Lenalidomide (CC-5013, Revlimid), a more potent drug with less neurotoxicity than thalidomide, has recently

been shown to have clinical activity in approximately 20% of patients (Cortes et al. 2005; Tefferi et al. 2005). It should be stressed however, that most patients will become refractory to medical therapies and consequently will require life-long transfusions with resulting iron overload.

15.7.1.6 Experimental Therapy

Etanercept, a recombinant form of the extracellular domain of tissue necrosis factor (TNF) receptor linked to the Fc fragment of human IgG, inhibits TNF- α , a key mediator of malignancy-associated fever, cachexia, and other constitutional symptoms. Two pilot studies have reported its use in CIMF. Steensma and colleagues (2002) observed a 60% reduction in severity of constitutional symptoms, while 20% experienced reduction of splenomegaly and/or improvement of cytopenias. Imatinib mesylate has been evaluated in CIMF on the basis that it inhibits the receptors PDGFR and KIT (Tefferi et al. 2002). However, the drug demonstrated limited efficacy and side effects led to the withdrawal in many patients. R115777, a farnesyl transferase inhibitor, has in vitro antiproliferative activity for CIMF progenitor cells. In a small study, R115777 produced improvement in anemia and splenomegaly in 25% of cases, with responses correlating with high VEGF levels (Cortes et al. 2003). Paradoxically, however, the VEGF tyrosine kinase inhibitor SU5416 possesses minimal therapeutic activity in CIMF (Giles et al. 2003). Studies evaluating the efficacy of new drugs including thalidomide analogues, proteasome inhibitors and VEGF neutralizing antibodies are currently underway.

15.7.2 Surgery and Radiotherapy

15.7.2.1 Splenectomy

The role of splenectomy in the management of myelofibrosis is now fairly well defined (Barosi et al. 1993; Mesa et al. 2004; Tefferi et al. 2000). In contrast to earlier reports, which suggested that the operation be performed in every patient at diagnosis, it is now clear that the procedure should be restricted to carefully selected cases with refractory hemolysis and/or thrombocytopenia, symptomatic splenomegaly, significant splenic infarction, and severe portal hypertension. Splenectomy does not prolong survival and even in the best units is asso-

ciated with morbidity and mortality rates of approximately 31% and 9%, respectively (Tefferi et al. 2000). The main postoperative complications include bleeding, thromboembolism, subphrenic abscess, and pulmonary atelectasis. In addition, compensatory hepatic myeloid metaplasia leading to rapid hepatic enlargement is an unusual but well-recognized complication, while an unexpectedly high rate of leukemic transformation has been documented (Barosi et al. 1998; López-Guillermo et al. 1991). The explanation for the increased blast transformation is unclear, but it is possible that the procedure could accelerate a pre-existing hyperproliferation state, especially as this complication has not been reported in healthy individuals. A significant postoperative thrombocytosis is observed in approximately 20% of patients and carries an increased thrombotic risk (Barosi et al. 1993a). Once a patient is considered a candidate for splenectomy, an extensive preoperative evaluation is required to determine if the cardiac, hepatic, renal, metabolic, and hemostatic risks are acceptable. The importance of the frequently associated coagulopathy needs to be stressed. Defective platelet aggregation is common and many patients have a prolonged bleeding time. In addition, some cases have laboratory evidence of disseminated intravascular coagulation, which may only become clinically apparent following surgical intervention, while others have a prolonged prothrombin time due to associated liver dysfunction, an isolated factor V deficiency, or the presence of circulating anticoagulants. Surgical expertise is essential, since the operation is often difficult as the spleen may be adherent to adjacent serosal surfaces as well as possessing numerous collateral vessels and dilated spleno-portal arteries and veins. Individuals operated on for portal hypertension and bleeding varices should have dynamic circulatory studies performed during the procedure, since portal hypertension due to splenomegaly is corrected by splenectomy (Silverstein and ReMine 1979), whereas cases secondary to intrahepatic obstruction require a portal-systemic shunt (Tefferi et al. 1994).

15.7.2.2 Radiotherapy

Radiotherapy should be considered as an alternative to splenectomy in those patients who are unfit for surgery. Several studies have reported symptomatic relief, with mild to moderate reduction in spleen size, which lasts for approximately 6 months (Bouabdallah et al. 2000; Elliot et al. 1998). A significant percentage of patients,

however, suffer unpredictable, life-threatening cytopenias, resulting in an overall mortality rate of 13%. The transient response, together with the high mortality rate for patients requiring subsequent splenomegaly, suggests that such therapy should not be regarded as an alternative to splenectomy in surgical candidates. Low-dose irradiation, however, remains the treatment of choice for peritoneal and pleural extramedullary hematopoiesis that results in ascites and pleural effusions, respectively (Bartlett et al. 1995; Leinweber et al. 1991), as well as for myeloid metaplasia of vital organs, including the lung, central nervous system, and liver (Price and Bell 1985; Steensma et al. 2002; Tefferi et al. 2001b).

15.7.3 Stem Cell Transplantation

15.7.3.1 Standard Allo-SCT

Currently, allogeneic hematopoietic stem cell transplantation (allo-SCT) is the only curative modality for patients with CIMF. Recently, two large studies have attempted to clarify the issues surrounding patient selection and outcome. Guardiola et al. (1999), in a retrospective multicenter study, reported the results of HLA-identical SCT in 55 patients (median age at transplantation 42 years, range 4–53). The 5-year probability of survival was $47\% \pm 8\%$ for the overall group, and $54\% \pm 8\%$ for patients receiving an unmanipulated HLA-matched related transplant. The 1-year probability of transplant-related mortality was $27\% \pm 6\%$. Hemoglobin (<10 g/dL), osteosclerosis, and a high-risk score at the time of transplantation were associated with a worse survival, whereas older age, karyotypic abnormalities, and lack of grade II–IV acute GVHD were associated with treatment failure. Deeg and colleagues (2003) reported the results in 56 patients treated at a single institution with conventional allo-SCT. The median age at transplantation was 43 years (range 10 to 66) with median disease duration of 33 months (range 3 to 312). Fifty-three patients achieved engraftment, while two died from relapse/progressive disease and 18 from other causes. The probability of surviving 3 years was 58%, with patients having lower Lille scores, higher platelet counts, less severe marrow fibrosis, and normal karyotypes doing better than those with more advanced disease. The role of pretransplant splenectomy is unclear. Guardiola and colleagues (1999) reported that although splenectomy was associated with a faster hematopoietic recovery, there was no associated survival benefit. A

pragmatic approach, therefore, in view of the mortality and morbidity of splenectomy, would be to restrict the procedure to patients where engraftment might be significantly delayed, for example in cases of osteomyelosclerosis or massive splenomegaly.

15.7.3.2 Reduced Intensity Allo-SCT

Evidence for a graft-versus-myelofibrosis (GvMy) effect has provided the rationale for exploring the role of reduced-intensity, or nonmyeloablative, allo-SCT in CIMF. Guardiola and colleagues (1999), for example, reported that absent or minimal GvHD in the context of conventional allo-SCT correlated with treatment failure, while the infusion of donor lymphocytes in patients failing allo-SCT may lead to disease eradication (Byrne et al. 2000; Cervantes et al. 2000). The initial reports of reduced-intensity allo-SCT were retrospective registry studies with patients receiving a range of conditioning regimes. Nevertheless, encouraging 1-year survival rates of 54% and 90% were reported (Hessling et al. 2002; Rondelli et al. 2003). Recently, the first prospective study using reduced-intensity conditioning has been published (Kröger et al. 2005). This series demonstrated that a busulphan- and fludarabine-based regimen, followed by allo-SCT from either a related or unrelated donor, is a feasible and effective therapeutic option with a low treatment-related mortality. Complete donor chimerism was seen in 95% of cases on day 100 with acute GvHD grades II–IV and III/IV occurring in 48% and 19% of cases respectively, while 55% developed chronic GvHD. The estimated overall and disease-free survival at 3 years was 84%, with no treatment failure, as defined by recurrence or progression of myelofibrosis, being observed after a median follow-up of 22 months. Importantly, despite *in vivo* T-cell depletion, no relapses were reported. In addition, the results from unrelated donors compared favorably with the outcome of the few cases of unrelated SCT reported using standard conditioning. In the largest series of the latter, for example, seven patients were transplanted from unrelated donors, but only one survived (Guardiola et al. 1999). It should be stressed, however, that the overall experience of reduced-intensity allo-SCT remains limited and crucial questions such as the impact of prior splenectomy, the influence of karyotype and the effect of the degree of fibrosis on outcome, remain unclear. As a result, the use of reduced-intensity conditioning should be restricted to patients aged 45–70 years with high-risk disease.

15.7.3.3 Autologous SCT

The risks of allo-SCT have led clinicians to investigate the palliative option of autologous-SCT, especially for the older patient or for those without a stem cell donor. The feasibility of this approach was demonstrated by Anderson and colleagues (2001) who reported a multicenter retrospective analysis of 21 patients, conditioned using single agent busulphan at a dose of 16 mg/kg. At 2 years the actuarial survival was 61%, with six patients having died of infection, graft failure, or progressive disease. Following transplantation, 15 of 21 patients showed evidence of significant clinical improvement that lasted up to 4 years, including ten of 17 cases with anemia, 7 of 10 patients with symptomatic splenomegaly, and 6 of 8 cases with thrombocytopenia. G-CSF priming was recommended since, although CIMF patients have high circulating CD34+ cell counts, most peripheral CD34+ cells are likely to lack the capacity to maintain long-term hematopoiesis. The mechanism for response is unclear but may include the restoration of intramedullary hematopoiesis as a result of reduced fibrosis, reduced sequestration of nonclonal cells following improvement in the degree of splenomegaly, and the restoration of normal hematopoiesis secondary to a reduction of malignant cells.

References

- Abu-Duhier FM, Goodeve AC, Care RS, Gari M, Wilson GA, Peake IR, Reilly JT (2003) Mutational analysis of class III receptor tyrosine kinases (C-KIT, c-FMS, FLT3) in idiopathic myelofibrosis. *Br J Haematol* 120:464–470
- Al-Assar O, Ul-Hassan A, Brown R, Wilson GA, Hammond DW, Reilly JT (2005) Gains on 9p are common genomic aberrations in idiopathic myelofibrosis: a comparative genomic hybridization study. *Br J Haematol* 129:66–71
- Aloe-Spiriti MA, Latagliata R, Avvisati G, Battistel V, Montevusco E, Spadea A, Petti MC (1993) Erythropoietin treatment of idiopathic myelofibrosis. *Haematol* 78:371–373
- Anderson JE, Tefferi A, Craig F, Holmberg L, Chauncey T, Appelbaum FR, Guardiola P, Callander N, Freytes C, Gazitt Y, Razvillas B, Deeg HJ (2001) Myeloablation and autologous peripheral blood stem cell rescue results in hematologic and clinical responses in patients with myelofibrosis. *Blood* 98:586–593
- Andreasson B, Swolin B, Kutti J (2002) Patients with idiopathic myelofibrosis show increased CD34+ cell concentrations in peripheral blood compared to patients with polycythaemia vera and essential thrombocythaemia. *Eur J Haematol* 68:189–193
- Altura RA, Head DR, Wang WC (2000) Long-term survival of infants with idiopathic myelofibrosis. *Br J Haematol* 109:459–462
- Apaja-Sarkkinen M, Autio-Harmanen H, Alavaikko M, Ristelli J, Risteli L (1986) Immunohistochemical study of basement membrane pro-

- teins and type III procollagen in myelofibrosis. *Br J Haematol* 63:571–580
- Arora B, Sirhan S, Hoyer JD, Mesa RA and Tefferi A (2004) Peripheral blood CD34 count in myelofibrosis with myeloid metaplasia: a prospective evaluation of prognostic value in 94 patients. *Br J Haematol* 128:42–48
- Ayyildiz O, Isikdogan A, Celik M, Muftuoglu E (2004) Intracranial meningeal extramedullary hematopoiesis inducing serious headache in a patient with idiopathic myelofibrosis. *J Pediatr Hematol Oncol* 26:28–29
- Badon SJ, Ansell J, Smith TW, Coslovsky R, Gill L, Woda BA (1985) Diabetes insipidus caused by extramedullary hematopoiesis. *Am J Clin Pathol* 83:509–512
- Balogh K, O'Hara CJ (1986) Myeloid metaplasia masquerading as a urethral caruncle. *J Urol* 135:789–790
- Barosi G, Berzuini C, Liberato LN, Costa A, Polino G, Ascarelli E (1988) A prognostic classification of myelofibrosis with myeloid metaplasia. *Br J Haematol* 70:397–401
- Barosi G, Ambrosetti A, Buratti A, Finelli C, Liberato NL, Quaglini S, Ricetti MM, Visani G, Tura S, Ascarelli E (1993a) Splenectomy for patients with myelofibrosis with myeloid metaplasia: pre-treatment variables and outcome prediction. *Leukemia* 7:200–206
- Barosi G, Liberato LN, Guarnone R (1993b) Serum erythropoietin in patients with myelofibrosis with myeloid metaplasia. *Br J Haematol* 83:365–369
- Barosi G, Ambrosetti A, Centra A, Falcone A, Finelli C, Foa P, Grossi A, Guarnone R, Rupoli S, Luciano L, Petti MC, Pogliani E, Russo D, Ruggeri M, Quaglini S (1998) Splenectomy and risk of blast transformation in myelofibrosis with myeloid metaplasia. *Blood* 91:3630–3636
- Barosi G, Ambrosetti A, Finelli C, Grossi A, Leoni P, Liberato NL, Petti MC, Pogliani E, Ricetti M, Rupoli S, Visani G, Tura S (1999) The Italian Consensus Conference on Diagnostic Criteria for Myelofibrosis with Myeloid Metaplasia. *Br J Haematol* 104:730–737
- Barosi G, Viarengo G, Pecci A, Rosti V, Piaggio G, Marchetti M, Frasson F (2001) Diagnostic and clinical relevance of the number of circulating CD34(+) cells in myelofibrosis with myeloid metaplasia. *Blood* 98:3249–3255
- Barosi G, Elliot M, Canepa L, Filippo B, Piccaluga PP, Visani G, Marchetti M, Pozzato G, Zorat F, Tefferi A (2002) Thalidomide in myelofibrosis with myeloid metaplasia: a pooled-analysis of individual patient data from 5 studies. *Leuk Lymphoma* 43:2301–2307
- Bartlett RP, Greipp PR, Tefferi A, Cupps RE, Mullan BP, Trastek VF (1995) Extramedullary hematopoiesis manifesting as a symptomatic pleural effusion. *Mayo Clin Proc* 70:1161–1164
- Baxter EJ, Scott LM, Campbell PJ, East C, Fouroudas N, Swanton S, Vassiliou GS, Bench AJ, Boyd EM, Curtin N, Scott MA, Erber WN, Green AR (2005) Acquired mutation of the tyrosine kinase JAK2 in human myeloproliferative disease. *Lancet* 365:1054–1061
- Bench AJ, Nacheva EP, Champion KM, Green AR (1998) Molecular genetics and cytogenetics of myeloproliferative disorders. *Baillieres Clin Haematol* 11:819–848
- Bernabei PA, Arcangeli A, Casini M, Grossi A, Padovani R, Ferrini PR (1986) Platelet-derived growth factor(s) mitogenic activity in patients with myeloproliferative disease. *Br J Haematol* 63:353–357
- Besa E, Nowell PC, Geller NL, Gardner FH (1982) Analysis of the androgen response of 23 patients with agnogenic myeloid metaplasia: the value of chromosome studies predicting response and survival. *Cancer* 49:308–313
- Bird GW, Wingham J, Richardson SG (1985) Myelofibrosis, autoimmune haemolytic anaemia and Tn-polyagglutinability. *Haematol* 18:99–103
- Bouabdallah R, Coso D, Gonzague-Casabianca L, Alzieu C, Resbeut M, Gastaut JA (2000) Safety and efficacy of splenic irradiation in the treatment of patients with idiopathic myelofibrosis: a report on 15 patients. *Leuk Res* 24:491–495
- Bourantas KL, Tsiara S, Christou L, Repousis P, Konstantinidou P, Bai M, Seferiadis K (1996) Combination therapy with recombinant human erythropoietin, interferon- α 2b and granulocyte-macrophage colony-stimulating factor in idiopathic myelofibrosis. *Acta Haematol* 96:79–82
- Brogi E, Wu T, Namiki A, Isner JM (1994) Indirect angiogenic cytokines upregulate VEGF and bFGF gene expression in vascular smooth muscle cells whereas hypoxia upregulates VEGF expression only. *Circulation* 90:649–652
- Brubaker LH, Briere J, Laszlo J, Kraut E, Landaw SA, Peterson P, Goldberg J, Donovan P (1982) Treatment of anaemia in myeloproliferative disorders: a randomized study of flouxymesterone v transfusions only. *Arch Intern Med* 142:1533–1537
- Buhr T, Buesche G, Choritz H, Langer F, Kreipe H (2003) Evolution of myelofibrosis in chronic idiopathic myelofibrosis as evidenced in sequential bone marrow biopsy specimens. *Am J Clin Pathol* 119:152–158
- Buschle M, Janssen JWG, Drexler H, Lyons J, Anger B, Bartram CR (1988). Evidence for pluripotent stem cell origin of idiopathic myelofibrosis: clonal analysis of a case characterised by a N-ras gene mutation. *Leukemia* 2:658–660
- Byrne JL, Beshti H, Clark D, Ellis I, Haynes AP, Das-Gupta E, Russell NH (1999) Induction of remission after donor leucocyte infusion for the treatment of relapsed chronic idiopathic myelofibrosis following allogeneic transplantation: evidence for a "graft vs myelofibrosis" effect. *Br J Haematol* 108:430–433
- Caligaris Cappio F, Vigiani R, Novarino A, Camussi G, Campana D, Gavosto F (1981) Idiopathic myelofibrosis: a possible role for immune-complexes in the pathogenesis of bone marrow fibrosis. *Br J Haematol* 49:17–21
- Cameron WR, Ronnert M, Brun A (1981) Extramedullary hematopoiesis of CNS in postpolycythemic myeloid metaplasia. *N Eng J Med* 305:765
- Campbell PJ, Griesshammer M, Dohmer K, Dohner R, Hasselbalch HC, Larsen TS, Pallisgaard N, Giraudier S, Le Bousse-Kerdiles MC, Desterke C, Guerton B, Dupriez B, Bordessoulle D, Harrison CN, Green AR, Reilly JT (2006) The V617F mutation in JAK2 is associated with poorer survival in idiopathic myelofibrosis. *Blood* 107:2098–2100
- Carlo-Stella C, Cazzola M, Gasner A, Barosi G, Dezza L, Meloni F, Pedrazzoli P, Hoelzer D, Ascarelli E (1987) Effects of recombinant alpha and gamma interferons on the in vitro growth of circulating hematopoietic progenitor cells (CFU-GEMM, CFU-Mk, BFU-E, and CFU-GM) from patients with myelofibrosis with myeloid metaplasia. *Blood* 70:1014–1019
- Castro-Malaspina H, Rabellino EM, Yen A, Nachman RL, Moore MAS (1981) Human megakaryocyte stimulation of proliferation of bone marrow fibroblasts. *Blood* 57:781–787
- Cervantes F, Pereira A, Esteve J, Rafel M, Cobo F, Rozman C, Montserrat E (1997) Identification of "short-lived" and "long-lived" patients at

- presentation of idiopathic myelofibrosis. *Br J Haematol* 97:635–640
- Cervantes F, Barosi G, Demory J-L, Reilly J, Guarone R, Dupriez B, Pereira A, Montserrat E (1998) Myelofibrosis with myeloid metaplasia in young individuals: disease characteristics, prognostic factors and identification of risk groups. *Br J Haematol* 102:684–690
- Cervantes F, Rovira M, Urbano-Ispizua A, Rozman M, Carreras E, Montserrat E (2000) Complete remission of idiopathic myelofibrosis following donor lymphocyte infusion after failure of allogeneic transplantation: demonstration of a graft-versus-myelofibrosis effect. *Bone Marrow Transplant* 26:697–699
- Cervantes F, Hernandez-Boluda JC, Alvarez A, Nadal E, Montserrat E (2002) Danazol treatment of idiopathic myelofibrosis with severe anaemia. *Haematol* 85:595–599
- Cervantes F, Alvarez-Larran A, Hernandez-Boluda JC, Sureda A, Torredadell M, Montserrat E (2004) Erythropoietin treatment of the anaemia of myelofibrosis with myeloid metaplasia: results in 20 patients and review of literature. *Br J Haematol* 127:399–403
- Cervantes F, Alvarez-Larran A, Domingo A, Arellano-Rodrigo E, Montserrat E (2005) Efficacy and tolerability of danazol as a treatment for the anaemia of myelofibrosis with myeloid metaplasia: long-term results in 30 patients. *Br J Haematol* 129:771–775
- Chaiter Y, Brenner B, Aghai E, Tatarsky I (1992) High incidence of myeloproliferative disorders in Ashkenazi Jews in northern Israel. *Leuk Lymphoma* 7:251–255
- Charbord P (1986) Increased vascularity of bone marrow in myelofibrosis. *Br J Haematol* 62:595–596
- Connolly TJ, Abruzzo JL, Schwab RH (1982) Agnogenic myeloid metaplasia with polyarteritis. *Journal of Rheumatology* 9:954–956
- Cornfield DB, Shipkin P, Alavi A, Becker J, Peyster R (1983). Intracranial myeloid metaplasia: diagnosis by CT and Fe52 scans and treatment by cranial irradiation. *Am J Hematol* 15:273–278
- Cortes J, Albitar M, Thomas D, Giles F, Kurzrock R, Thibault A, Rackoff W, Koller C, O'Brien S, Garcia-Mancro G, Talpaz M, Kantarjian H (2003) Efficacy of the farnesyl transferase inhibitor R115777 in chronic myeloid leukemia and other hematologic malignancies. *Blood* 101:1692–1697
- Cortes J, Thomas D, Verstovsek S, Giles F, Beran M, Koller C, Kantarjian H (2005) Phase II study of Lenalidomide (CC-5013, Revlimid) for patients (pts) with myelofibrosis (MF) (abstract) 106:114a
- Cox MC, Panetta P, Venditti A, Abruzzese E, Del Poeta G, Cantonette M, Amadori S (2001) New reciprocal translocation t(6;10) (q27;q11) associated with idiopathic myelofibrosis. *Leuk Res* 25:349–351
- Craig JIO, Anthony RS, Parker AC (1991) Circulating progenitor cells in myelofibrosis: the effect of recombinant $\alpha 2b$ interferon in vivo and in vitro. *Br J Haematol* 78:155–160
- Cunietti E, Gandini R, Marcaro G et al (1981) Defective platelet aggregation and increased platelet turnover in patients with myelofibrosis and other myeloproliferative diseases. *Scand J Haematol* 26:339–344
- Dalley A, Smith JM, Reilly JT, MacNeil S (1996) Investigation of calmodulin and basic fibroblast growth factor (bFGF) in idiopathic myelofibrosis: evidence for a role of extracellular calmodulin in fibroblast proliferation. *Br J Haematol* 93:856–862
- Dameshek W (1951) Some speculations on the myeloproliferative syndromes. *Blood* 6:372–375
- Deeg HJ, Golley TA, Flowers ME, Sale GE, Slattery JT, Anasetti C, Chauncey TR, Doney K, Georges GE, Kiem HP, Martin PJ, Petersdorf EW, Radich J, Sanders JE, Sandmaier BM, Warren EH, Witherspoon RP, Storb R, Appelbaum FR (2003) Allogeneic hematopoietic stem cell transplantation for myelofibrosis. *Blood* 102:3912–3918
- Demory JL, Dupriez B, Fenaux P, Lai JL, Beuscart R, Jouet JP, Deminatti M, Bauters F (1988) Cytogenetic studies and their prognostic significance in agnogenic myeloid metaplasia: a report on 47 cases. *Blood* 72:855–859
- Den Ottolander GJ, Te velde J, Brederoo P, Geraedts JPM, Snee TH, Willmze R, Zwaan FE, Haak HI, Muller HP, Bieger R (1979) Megakaryoblastic leukaemia (acute myelofibrosis): a report of three cases. *Br J Haematol* 42:9–20
- Dingli D, Grand FH, Mahaffey V, Spurbeck J, Ross FM, Reilly JT, Cross NCP, Dewald GW, Tefferi A (2005) Der(6)t(1;6)(q21–23; p21.3): the first specific cytogenetic abnormality in myelofibrosis with myeloid metaplasia. *Br J Haematol* 130:229–232
- Dolan G, Forrest PL, Eastham JM, Reilly JT (1991) Reduced platelet PDGF levels in idiopathic myelofibrosis. *Br J Haematol* 78:586–588
- Donti E, Tabilio A, Bocchini F et al (1990) Partial trisomy 1q in idiopathic myelofibrosis. *Leuk Res* 14:1035–1040
- Dougan LE, Matthews MLV, Armstrong BK (1981) The effect of diagnostic review on the estimated incidence of lymphatic and haematopoietic neoplasms in Western Australia. *Cancer* 48:866–872
- Dupriez B, Morel P, Demory JL, Lai JL, Simon M, Plantier I, Bautiers F (1996) Prognostic factors in agnogenic myeloid metaplasia: a report on 195 cases with a new scoring system. *Blood* 88:1013–1018
- Eastham JM, Reilly JT, MacNeil S (1994) Raised urinary calmodulin levels in idiopathic myelofibrosis: possible implications for the aetiology of fibrosis. *Br J Haematol* 86:668–670
- Elliot MA, Chen MG, Silverstein MN, Tefferi A (1998) Splenic irradiation for symptomatic splenomegaly associated with myelofibrosis with myeloid metaplasia. *Br J Haematol* 103:505–511
- Emadi S, Clay D, Desterke C, Guerton B, Maquarrie E, Charpentier A, Jamin C and Le Bousse-Kerdiles, M-C (2005) IL-8 and its CXCR1 and CXCR2 receptors participate in the control of megakaryocytic proliferation, differentiation, and ploidy in myeloid metaplasia with myelofibrosis. *Blood* 105:464–473
- Faoro LN, Tefferi A, Mesa R (2005) Long-term analysis of the palliative benefit of 2-chlorodeoxyadenosine for myelofibrosis with myeloid metaplasia. *Eur J Haematol* 74:117–120
- Fava RA, Casey TT, Wilcox J, Pelton RW, Moses HL, Nannery LB (1990) Synthesis of transforming growth factor- $\beta 1$ by megakaryocytes and its localization to megakaryocytes and platelet α -granules. *Blood* 76:1946–1955
- Gaidano G, Guerrasio A, Serra A et al (1993) Mutations in the p53 and ras family genes are associated with tumour progression of bcr/abl negative chronic myeloproliferative disorders. *Leukemia* 7: 946–953
- Garcia S, Miguel A, Linares M, Navarro M, Colomina P (1989) Idiopathic myelofibrosis terminating in erythroleukemia. *Am J Hematol* 32:70–71
- Gay S, Gay RE, Prchal JT (1984) Immunohistological studies of bone marrow collagen. In: Berk P, Castro-Malaspin H, Wasserman LR (eds) *Myelofibrosis and the Biology of Connective Tissue*. Liss, New York, pp 291–306
- Geogii A, Buesche G, Kreft A (1998) The histopathology of chronic myeloproliferative diseases. *Baillieres Clin Haematol* 11:721–749

- Gersuk G, Carmel R, Pattengale PK (1989) Platelet-derived growth factor concentrations in platelet-poor and urine from patients with myeloproliferative disorders. *Blood* 74:2330–2334
- Gibson LE, Dicken CH, Flach DB (1985) Neutrophilic dermatoses and myeloproliferative disease: report of two cases. *Mayo Clin Proc* 60:735–740
- Gilbert HS, Praloran V, Stanley ER (1989) Increased CSF-1 (M-CSF) in myeloproliferative disease: association with myeloid metaplasia and peripheral bone marrow extension. *Blood* 74:1231–1234
- Giles FJ, Cooper MA, Silverman L, Karp JE, Lancet JE, Zangari M, Shami PJ, Khan KD, Hannah AL, Cherrington JM, Thomas DA, Garcia-Manero G, Albitar M, Kantarjian HM, Stopeck AT (2003) Phase II study of SU5416 – a small-molecule, vascular endothelial growth factor tyrosine-kinase receptor inhibitor – in patients with refractory myeloproliferative diseases. *Cancer* 97:1920–1928
- Giraudier S, Chagraoui H, Komura E et al (2002) Overexpression of FKPBP1 in idiopathic myelofibrosis regulates the growth factor independence of megakaryocyte progenitors. *Blood* 100:2932–2940
- Gordon BR, Colman M, Kohen P, Day NK (1981) Immunologic abnormalities in myelofibrosis with activation of the complement system. *Blood* 58:904–910
- Guardiola P, Anderson JE, Bandini G, Cervantes F, Runde V, Arcese W, Bacigalupo A, Przepiorka D, O'Donnell MR, Polchi P, Buzyn A, Sutton L, Cazals-Hatem D, Sale G, de Witte T, Deeg HJ, Gluckman E (1999) Allogeneic stem cell transplantation for agnogenic myeloid metaplasia: a European Group for Blood and Bone Marrow Transplantation, Société Française de Greffe de Moelle, Gruppo Italiano per il Trapianto del Midollo Osseo, and Fred Hutchinson Cancer Centre Research Center Collaborative Study. *Blood* 93:2831–2838
- Han ZC, Briere J, Nedellec G, Abgrall JF, Senzebe L, Parent D, Guern G (1988) Characteristics of circulating megakaryocyte progenitor (CFU-MK) in patients with myelofibrosis. *Eur J Haematol* 40:130–135
- Harmey JH, Dimitriadis E, Kay E, Redmond HP, Bouchier-Hayes D (1998) Regulation of macrophage production of vascular endothelial growth factor (VEGF) by hypoxia and transforming growth factor β -1. *Ann Surg Oncol* 5:271–278
- Hasselbalch H, Junker P, Lisse I, Bentsen KD, Risteli L, Risteli J (1986) Serum markers for type IV collagen and type III procollagen in the myelofibrosis-osteomyelosclerosis syndrome and other chronic myeloproliferative disorders. *Am J Hematol* 23:101–111
- Hasselbalch HC, Clausen NT, Jensen BA (2002) Successful treatment of anaemia in idiopathic myelofibrosis with recombinant human erythropoietin. *Am J Haematol* 70:92–99
- Hast R, Engstedt L, Jameson S, Killander A, Lundh B, Reizenstein P, Skarberg KO, Uden AM, Wadman B (1978) Oxymethalone treatment in myelofibrosis. *Blut* 37:12–26
- Hernandez JM, San Miguel JF, Gonzalez M et al (1992) Development of acute leukemia after idiopathic myelofibrosis. *J Clin Pathol* 45:427–430
- Hessling J, Kroger N, Werner M, Zabelina T, Hansen A, Kordes U, Ayuk FA, Renges H, Panse J, Erttmann R, Zander AR (2002) Dose-reduced conditioning regimen followed by allogeneic stem cell transplantation in patients with myelofibrosis with myeloid metaplasia. *Br J Haematol* 119:769–772
- Hibbin JA, Njoku OS, Matutes E, Lewis SM, Goldman JM (1984) Myeloid progenitor cells in the circulation of patients with myelofibrosis and other myeloproliferative disorders. *Br J Haematol* 57:495–503
- Horwood E, Dowson H, Gupta R, Kaczmarek R, Williamson M (2003) Myelofibrosis presenting as spinal cord compression. *J Clin Pathol* 56:154–156
- Hu H (1987) Benzene-associated myelofibrosis. *Annals of Internal Medicine* 106:171–173
- Humphrey PA, Vollmer RT (1991) Extramedullary hematopoiesis in the prostate. *Am J Surg Pathol* 15:486–490
- Iványi JL, Mahunka M, Papp A, Kiss A, Telek B (1984) Prognostic significance of bone marrow reticulin fibres in idiopathic myelofibrosis: evaluation of clinicopathological parameters in a scoring system. *Haematol* 26:75–86
- Jack FR, Smith SR, Saunders PWG (1994) Idiopathic myelofibrosis: anaemia can respond to low-dose dexamethasone. *Br J Haematol* 87:876–884
- Jacobson RJ, Salo A, Fialkow PJ (1978) Agnogenic myeloid metaplasia: a clonal proliferation of haematopoietic stem cells with secondary myelofibrosis. *Blood* 51:189–194
- James C, Ugo V, Le Couedic JP, Staerk J, Delmonneau F, Lacout C, Garcon L, Raslova H, Berger R, Bennaceur-Griscelli A, Villeval JL, Constantinescu SN, Casadevall N, Vainchenker W (2005) A unique clonal JAK2 mutation leading to constitutive signalling causes polycythaemia vera. *Nature* 434:1144–1148
- Jantunen E, Hänninen A, Naukkarinen A, Vornanen M, Lahtinen R (1994) Gray platelet syndrome with splenomegaly and signs of extramedullary hematopoiesis: case report with review of literature. *Am J Hematol* 46:218–224
- Johnson JB, Dalal BI, Israels SJ, Oh S, McMillan E, Begleiter A, Michaud G, Israels LG, Greenberg AH (1995) Deposition of transforming growth factor- β in the marrow in myelofibrosis, and the intracellular localization and secretion of TGF- β by leukemic cells. *Am J Clin Pathol* 103:574–582
- Jones LC, Tefferi A, Idos GE, Kumagai T, Hofman WK, Koeffler HP (2004) RARbeta2 is a candidate suppressor gene in myelofibrosis with myeloid metaplasia. *Oncogene* 23:7846–7853
- Jones LC, Tefferi A, Vuong PT, Desmond JC, Hofmann WK, Koeffler HP (2005) Detection of aberrant gene expression in CD34+ hematopoietic stem cells from patients with agnogenic myeloid metaplasia using oligonucleotide microarrays. *Stem Cells* 23:631–637
- Jowitt SN, Burke DK, Leggat HM, Lewis PS, Cryer RJ (1997) Pleural effusions secondary to extramedullary hematopoiesis in a patient with idiopathic myelofibrosis responding to pleurodesis and hydroxyurea. *Clin Lab Haematol* 19:283–285
- Kaelin WG, Spivak JL (1986) Systemic lupus erythematosus and myelofibrosis. *Am J Med* 81:935–938
- Kahn A, Bernard JF, Cottreau D, Marie J, Boivin P (1975) Gd(–) Abrami: a deficient G-6PD variant with hemizygous expression in blood cells of a woman with primary myelofibrosis. *Humangenetik* 30:41–46
- Katoh O, Kimura A, Kuramoto A (1988) Platelet-derived growth factor is decreased in patients with myeloproliferative disorders. *Am J Hematol* 27:276–280
- Kobayashi S, Teramura M, Hoshino S, Motoji T, Oshimi K, Mizoguchi H (1993) Circulating megakaryocyte progenitors in myeloproliferative disorders are hypersensitive to interleukin-3. *Br J Haematol* 83:539–544

- Komura E, Chagraoui H, de Mas VM, Blanchet B, de Sepulveda P, LARBRET F, Larghero J, Tulliez M, Debili N, Vainchenker W, Giraudier S (2003) Spontaneous STAT5 activation induces growth factor independence in idiopathic myelofibrosis: possible relationship with FKBP51 overexpression. *Exp Hematol* 31:622–630
- Komura E, Tonetti C, Penard-Lacronique V, Chagraoui H, Lacout C, Le-couedic JP, Rameau P, Debili N, Vainchenker W, Giraudier S (2005) Role for the nuclear factor kappaB pathway in transforming growth factor- β 1 production in idiopathic myelofibrosis: possible relationship with FK506 binding protein 51 overexpression. *Cancer Res* 65:3281–3289
- Kralovics R, Passamonti F, Buser AS, Teo SS, Tiedt R, Passweg JR, Tichelli A, Cazzola M, Skoda RC (2005) A gain-of-function mutation of JAK2 in myeloproliferative disorders. *N Engl J Med* 352:1779–1790
- Kreft A, Weiss M, Wiese B, Choritz H, Buhr T, Busche G, Georgii A (2003) Chronic idiopathic myelofibrosis: prognostic impact of myelofibrosis and clinical parameters on event-free survival in 122 patients who presented in prefibrotic and fibrotic stages. A retrospective study identifying subgroups of different prognoses by using the RECPAM method. *Ann Hematol* 82:605–611
- Kreipe H, Jaquet K, Felgner J, Radzum HJ, Parwaresch MR (1991) Clonal granulocytes and marrow cells in the cellular phase of agnogenic myeloid metaplasia. *Blood* 78:1814–1817
- Kröger N, Zabelina T, Scheider H, Panse J, Ayuk F, Stute N, Fehse N, Waschke O, Fehse B, Kvasnicka HM, Thiele J, Zander A (2005) Pilot study of reduced-intensity conditioning followed by allogeneic stem cell transplantation from related and unrelated donors in patients with myelofibrosis. *Br J Haematol* 128:690–697
- Kupferschmid JP, Shahian DM, Villanueva AG (1993) Massive hemothorax associated with intrathoracic extramedullary hematopoiesis involving the pleura. *Chest* 103:974–975
- Kvasnicka HM, Thiele J, Werden C, Zankovich R, Diehl V, Fischer R (1997) Prognostic factors in idiopathic (primary) osteomyelofibrosis. *Cancer* 80:708–719
- Kvasnicka HM, Thiele J, Regn C, Zankovich R, Diehl V, Fischer R (1999) Prognostic impact of apoptosis and proliferation in idiopathic (primary) myelofibrosis. *Ann Hematol* 78:65–72
- Kvasnicka HM, Thiele J, Schroeder M, von Loesch C, Diehl V (2004) Bone marrow angiogenesis – methods of quantification and changes evolving in chronic myeloproliferative disorders. *Histol Histopathol* 19:1245–1260
- Landolfi R, Colosimo C, De Candia E, Castellana MA, De Cristofara R, Trodella L, Leone G (1988) Meningeal hematopoiesis causing exophthalmos and hemiparesis in myelofibrosis: effect of radiotherapy. *Cancer* 62:2346–2349
- Le Bousse-Kerdilès M-C, Chevillard S, Charpentier A, Romquin N, Clay D, Smadja-Jaffe F, Praloran V, Dupriez B, Demory J-L, Jasmin C, Martyr M-C (1996) Differential expression of transforming growth factor- β , basic fibroblast growth factor, and their receptors in CD34⁺ hematopoietic progenitor cells from patients with myelofibrosis and myeloid metaplasia. *Blood* 88:4534–4546
- Leinweber C, Order SE, Calkins AR (1991) Whole-abdominal irradiation for the management of gastrointestinal and abdominal manifestations of agnogenic myeloid metaplasia. *Cancer* 68:1251–1254
- Leoni P, Ruploi S, Salvi A, Sambo P, Cinciripini A, Gabriella A (1993) Antibodies against terminal galactosyl α (1–3) galactose epitopes in patients with idiopathic myelofibrosis. *Br J Haematol* 85:313–319
- Levine RL, Wadleigh M, Cools J, Ebert BL, Wernig G, Huntly BJP, Boggon TJ, Wlodarska I, Clark JJ, Moore S, Adelsperger J, Koo S, Lee JC, Gabriel S, Mercher T, D'Andrea A, Fröhling S, Döhner K, Marynen P, Vandenbergh P, Mesa RA, Tefferi A, Griffin JD, Eck MJ, Sellers WR, Meyerson M, Golub TR, Lee SJ, Gilliland DG (2005) Activating mutation in the tyrosine kinase JAK2 in polycythemia vera, essential thrombocythemia and myeloid metaplasia with myelofibrosis. *Cancer Cell* 7:387–397
- Levy V, Bourgarit A, Delmer A, Legrand O, Baudard M, Rio B, Zittoun R (1996) Treatment of agnogenic myeloid metaplasia with danazol: a report of four cases. *Am J Haematol* 53:239–241
- Lewis CM, Pegrum GD (1972) Immunocomplexes in myelofibrosis: a possible guide to management. *Br J Haematol* 39:233–239
- Ligumski M, Polliack A, Benbassat J (1979) Metaplasia of the central nervous system in patients with myelofibrosis and agnogenic myeloid metaplasia: report of 3 cases and review of literature. *Am J Med Sci* 275:99–103
- Loewy G, Mathew A, Distenfeld A (1994) Skin manifestations of agnogenic myeloid metaplasia. *Am J Hematol* 45:167–170
- Lofvenberg E, Wahlin A, Roos G, Ost A (1990) Reversal of myelofibrosis by hydroxyurea. *Eur J Haematol* 44:33–38
- López-Guillermo A, Cervantes F, Bruguera M, Pereira A, Feliú E, Rozman C (1991) Liver dysfunction following splenectomy in idiopathic myelofibrosis: a study of 10 patients. *Acta Haematol* 85:184–188
- Lundh B, Brandt L, Cronqvist S, Eyrich R (1982) Intracranial myeloid metaplasia in myelofibrosis. *Scand J Haematol* 28:91–94
- Macdonald DHC, Lahiri D, Chase A, Sohal J, Goldman JM, Cross NCP (1999) A case of myelofibrosis with a t(4;13)(q25;q12): evidence for involvement of a second 13q12 locus in chronic myeloproliferative disorders. *Br J Haematol* 105:771–774
- Mackinnon S, McNicol AM, Lee FD, McDonald GA (1986) Myelofibrosis complicated by intestinal extramedullary haematopoiesis and acute bowel obstruction. *J Clin Pathol* 39:677–679
- Manoharan A (1991) Management of myelofibrosis with intermittent hydroxyurea. *Br J Haematol* 77:252–254
- Manoharan A, Pitney WR (1984) Chemotherapy resolves symptoms and reverses marrow fibrosis in myelofibrosis. *Scand J Haematol* 33:453–459
- Marchetti M, Barosi G, Balestri F, Viarengo G, Gentili S, Barulli S, Demory J-L, Ilariucci F, Volpe A, Bordessoule D, Grossi A, Le Bousse-Kerdilès MC, Caenazzo A, Pecci A, Falcone A, Broccia G, Bendotti C, Bauduer F, Buccisano F, Dupriez B (2004) Low-dose thalidomide ameliorates cytopenias and splenomegaly in myelofibrosis with myeloid metaplasia: a phase II trial. *J Clin Oncol* 22:424–431
- Martyr M-C, Magdelenat H, Bryckaert MC, Iain-Bidron C, Calvo F (1991) Increased intraplatelet levels of platelet-derived growth factor and transforming growth factor- β in patients with myelofibrosis and myeloid metaplasia. *Br J Haematol* 77:80–86
- Martyr M-C, Le Bousse-Kerdilès M-C, Romquin N, Chevillard S, Praloran V, Demory J-L, Dupriez B (1997) Elevated levels of basic growth factor in megakaryocytes and platelets from patients with idiopathic myelofibrosis. *Br J Haematol* 97:441–448
- Mesa RA, Silverstein MN, Jacobsen SJ, Wollan PC, Tefferi A (1997) Population-based incidence and survival figures in essential thrombocythemia and agnogenic myeloid metaplasia: an Olmstead County Study 1976–1995. *Am J Hematol* 61:10–15

- Mesa MA, Hanson CA, Rajkumar VS, Schroeder G, Tefferi A (2000) Evaluation and clinical correlations of bone marrow angiogenesis in myelofibrosis with myeloid metaplasia. *Blood* 96:3374–3380
- Mesa RA, Steensma DP, Pardanani A, Li, C-Y, Elliot M, Kaufmann SH, Wiseman G, Gray LA, Schroeder G, Reeder T, Zeldis JB, Tefferi A (2003) A phase 2 trial of combination low-dose thalidomide and prednisone for the treatment of myelofibrosis with myeloid metaplasia. *Blood* 101:2534–2541
- Mesa R, Li C-Y, Ketterling RP, Schroeder GS, Knudson RA, Tefferi A (2005) Leukemic transformation in myelofibrosis with myeloid metaplasia: a single-institution experience with 91 cases. *Blood* 105:973–977
- Murate T, Yamashita K, Isogai C, Suzuki H, Ichihara M, Hatano S, Nakahara Y, Kinoshita T, Nagasaka T, Yoshida S, Komatsu N, Miura Y, Hotta T, Fujimoto N, Saito H, Hayakawa T (1997) The production of tissue inhibitors of metalloproteinases (TIMPS) in megakaryopoiesis: possible role of platelet- and megakaryocyte-derived TIMPS in bone marrow fibrosis. *Br J Haematol* 99:181–189
- Nakahata J, Takahashi M, Fuse I, Nakamori Y, Nomoto N, Saitoh H, Tatewaki W, Imanari A, Takeshige T, Koike T (1993) Paroxysmal nocturnal hemoglobinuria with myelofibrosis: progression to acute myeloblastic leukemia. *Leuk Lymphoma* 12:137–142
- Njoku OS, Lewis SM, Catovsky D, Gordon-Smith EC (1983) Anaemia in myelofibrosis: its value in prognosis. *Br J Haematol* 54:70–89
- Novetsky A, Wang JC, Chen C (1997) Plasma VEGF levels in patients with primary myelofibrosis and other myeloproliferative disorders. *Blood* 90:(abstract)
- O'Mahony CA, Albo D, Tuszyński GP, Berger DH (1998) Transforming growth factor-beta 1 inhibits generation of angiostatin by human pancreatic cancer cells. *Surgery* 124:388–393
- Overall CM, Wrana JL, Sodek J (1989) Independent regulation of collagenase, 72 kDa-progelatinase and metalloendoproteinase inhibitor (TIMP) expression in human fibroblasts by transforming growth factor- β . *J Biol Chem* 264:1860–1869
- Passamonti F, Vanelli L, Malabarba L, Rumi E, Pungolino E, Malcovati L, Pascutto C, Morra E, Lazzarino M, Cazzola M (2003) Clinical utility of the absolute number of circulating CD34-positive cells in patients with chronic myeloproliferative disorders. *Haematol* 88:1123–1129
- Perianin A, Labro-Bryskier MT, Marquetty C et al (1984) Glutathione reductase and nitro-blue tetrazolium reduction deficiencies in neutrophils of patients with primary idiopathic myelofibrosis. *Clin Exp Immunol* 57:244–248
- Petti MC, Latagliata R, Spadea T, Spadea A, Montefusco E, Aloe Spiriti MA, Avvisati G, Breccia M, Pescarmona E, Mandelli F (2002) Melphalan treatment in patients with myelofibrosis with myeloid metaplasia. *Br J Haematol* 116:576–581
- Pietrasanta D, Clavio M, Vallebella E, Beltrami G, Cavaliere M, Gobbi M (1997) Long-lasting effect of cyclosporin-A on anaemia associated with idiopathic myelofibrosis. *Haematol* 82:458–459
- Polliak A, Prokocimer M, Matzner Y (1980) Lymphoblastic leukemic transformation (lymphoblastic crisis) in myelofibrosis and myeloid metaplasia. *Am J Hematol* 9:211–220
- Price F, Bell H (1985) Spinal cord compression due to extramedullary hematopoiesis. Successful treatment in a patient with long standing myelofibrosis. *JAMA* 253:2876–2877
- Rameshwar P, Denny TN, Stein D, Gascon P (1994) Monocyte adhesion in patients with bone marrow fibrosis is required for the production of fibrogenic cytokines. Potential role for interleukin-1 and TGF- β . *J Immunol* 153:2819–2830
- Rameshwar P, Chang VT, Gascon P (1996) Implication of CD44 in adhesion-mediated overproduction of TGF- β and IL-1 in monocytes from patients with bone marrow fibrosis. *Br J Haematol* 96:22–29
- Reeder TL, Bailey RJ, Dewald GW, Tefferi A (2003) Both T and B lymphocytes may be clonally involved in myelofibrosis with myeloid metaplasia. *Blood* 101:1981–1983
- Reilly JT (2005) Cytogenetic and molecular genetic abnormalities in agnogenic myeloid metaplasia. *Sem Oncol* 32:359–364
- Reilly JT, Nash JGR, Mackie MJ, McVerry BA (1985 a) Immuno-enzymatic detection of fibronectin in normal and pathological haematopoietic tissue. *Br J Haematol* 59:497–504
- Reilly JT, Nash JGR, Mackie MJ, McVerry BA (1985 b) Endothelial cell proliferation in myelofibrosis. *Br J Haematol* 60:625–630
- Reilly JT, Nash JGR (1988) Vitronectin (serum spreading factor): its localization in normal and fibrotic tissue. *J Clin Pathol* 59:1269–1272
- Reilly JT, Barnett D, Dolan G, Forrest P, Eastham J, Smith A (1993) Characterization of an acute micromegakaryocytic leukaemia: evidence for the pathogenesis of myelofibrosis. *Br J Haematol* 83:58–62
- Reilly JT, Wilson G, Barnett D, Watmore A, Potter A (1994) Karyotypic and ras gene mutational analysis in idiopathic myelofibrosis. *Br J Haematol* 88:575–581
- Reilly JT, Brindley L, Kay M, Fielding S, Kennedy A, Dolan G, Smith A (1995) Bone marrow and serum connective tissue polypeptides in idiopathic myelofibrosis. *Clin Lab Haematol* 17:35–39
- Reilly JT, Snowden JA, Spearing RL, Fitzgerald PM, Jones N, Watmore A, Potter A (1997) Cytogenetic abnormalities and their prognostic significance in idiopathic myelofibrosis: a study of 106 cases. *Br J Haematol* 98:96–102
- Rege-Cambrin G, Speleman F, Kerim S, Sacravaglio P, Carozzi F, Dal Cin P, Michaux JL, Offner F, Saglio G, Van den Berghe H (1991) Extra translocation +der (1q9p) is a prognostic indicator in myeloproliferative disorders. *Leukemia* 5:1059–1063
- Roberts AB, Sporn MB, Assoian RK, Smith MJ, Roche NS, Wakefield LM, Heine UI, Liotta LA, Falanga V, Kehr JH, Fauci AS (1986) Transforming growth factor- β : rapid induction of fibrosis and angiogenesis in vivo and stimulation of collagen formation in vitro. *Proc Nat Acad Sci USA* 83:4167–4171
- Rodríguez JN, Martino ML, Diéguez JC, Prados D (1998) RhuEpo for the treatment of anaemia in myelofibrosis with myeloid metaplasia. Experience in 6 patients and meta-analytical approach. *Haematol* 83:616–621
- Romano M, Viero P, Cortellazzo S, Barbui T, Donati MB, Poggi A (1990) Platelet-derived mitogenic activity and bone marrow fibrosis in myeloproliferative disorders. *Haemostasis* 20:162–168
- Rondeau E, Soal-Celigny P, Dhermy D, Wraclans M, Brousse N, Bernard JF, Boivin P (1983) Immune disorders in agnogenic myeloid metaplasia: relation to fibrosis. *Br J Haematol* 53:467–475
- Rondelli D (2003) Non-myeoablative allogeneic HSCT in high-risk patients with myelofibrosis. *Blood* 102
- Rozman C, Giralt M, Feliu E, Rubio D, Cortes, M-T (1991) Life expectancy of patients with chronic nonleukemic myeloproliferative disorders. *Cancer* 67:2658–2663
- Rupoli S, Da Lio L, Sisti S et al (1994) Primary myelofibrosis: a detailed statistical analysis of the clinicopathological variables influencing survival. *Ann Hematol* 68:205–212

- Sagaster V, Jager E, Weltermann A, Schwarzingler I, Gisslinger H, Lechner K, Geissler K, Oehler L (2003) Circulating hematopoietic progenitor cells predict survival in patients with myelofibrosis and myeloid metaplasia. *Hematologica* 88:1204–1212
- Sacchi S, Curci G, Piccinini L, Messeroti A, Cucci F, Bursi R, Zaniol P, Torrelli V (1986) Platelet alpha-granule release in chronic myeloproliferative disorders with thrombocytosis. *J Clin Lab Invest* 46:163–166
- Schäfer AI (1982) Deficiency of platelet lipoxygenase activity in myeloproliferative disorders. *N Eng J Med* 306:381–386
- Schmitt A, Drouin A, Masse JM, Guichard J, Shagraoui H, Cramer EM (2002) Polymorphonuclear neutrophil and megakaryocyte mutual involvement in myelofibrosis pathogenesis. *Leuk Lymphoma* 43:719–724
- Schnuelle P, Waldherr R, Lehmann KJ, Woenckhaus J, Back W, Niemir Z, van der Woude FJ (1999) Idiopathic myelofibrosis with extramedullary hematopoiesis in the kidneys. *Clin Nephrol* 52:256–262
- Shaheen SP, Talwalker SS, Simons R, Yam L (2005) Acute lymphoblastic leukemic transformation in a patient with chronic idiopathic myelofibrosis and paroxysmal nocturnal hemoglobinuria: a case report and review of literature. *Arch Pathol Lab Med* 129:96–99
- Sharma BK, Pounder RE, Cruse JP, Knowles SM, Lewis AA (1986) Extramedullary hematopoiesis in the small bowel. *Gut* 27:873–875
- Silverstein MN (1975) Agnogenic myeloid metaplasia. Publishing Sciences Groups, Massachusetts, p 94–95
- Silverstein MN, ReMine WH (1979) Splenectomy in myeloid metaplasia. *Blood* 53:515–518
- Sinclair EJ, Forrest EC, Reilly JT, Watmore AE, Potter AM (2002) Fluorescence in situ hybridization analysis of 25 cases of idiopathic myelofibrosis and two cases of secondary myelofibrosis: monoallelic loss of RB1, D13S319 and D13S25 loci associated with cytogenetic deletion and translocation involving 13q14. *Br J Haematol* 113:365–368
- Steensma DP, Hook CC, Stafford SL, Tefferi A (2002) Low-dose, single fraction, whole-lung radiotherapy for pulmonary hypertension associated with myelofibrosis and myeloid metaplasia. *Br J Haematol* 118:813–816
- Strasser-Weippl K, Steurer M, Kees M, Augustin F, Tzankov A, Dirnhofer S, Fiegl M, Gisslinger H, Zofer N, Ludwig H (2005) Chromosome 7 deletions are associated with unfavorable prognosis in myelofibrosis with myeloid metaplasia. *Blood* 105:4146
- Sugimoto N, Ishikawa T, Gotoh S, Shinzato I, Matsusita A, Nagai K, Ohgoh N, Takahashi T (2004) Primary myelofibrosis terminated in basophilic leukemia and successful allogeneic bone marrow transplantation. *Int J Hematol* 80:183–185
- Taksin AL, Couedic JP, Dusanter-Fourt I, Masse A, Giraudier S, Katz A, Wendling F, Vainchenker W, Casadevall N, Debili N (1999) Autonomous megakaryocyte growth in essential thrombocythemia and idiopathic myelofibrosis is not related to a c-mpl mutation or to an autocrine stimulation by Mpl-L. *Blood* 93:125–139
- Tefferi A, Silverstein MN (1994) Recombinant human erythropoietin therapy in patients with myelofibrosis with myeloid metaplasia (letter). *Br J Haematol* 86:893–896
- Tefferi A, Barrett SM, Silverstein MN, Nagorney DM (1994) Outcome of portal-systemic shunt surgery for portal hypertension associated with intrahepatic obstruction in patients with agnogenic myeloid metaplasia. *Am J Hematol* 46:325–328
- Tefferi A, Silverstein MN, Li C-Y (1997) 2-chlorodeoxyadenosine treatment after splenectomy in patients who have myelofibrosis with myeloid metaplasia. *Br J Haematol* 99:352–357
- Tefferi A, Mesa RA, Nagomey DN, Schroeder G, Silverstein MN (2000) Splenectomy in myelofibrosis with myeloid metaplasia: a single-institution experience with 223 patients. *Blood* 95:2226–2233
- Tefferi A, Mesa RA, Schroeder G, Hanson CA, Li CY, Dewald GW (2001 a) Cytogenetic findings and their clinical relevance in myelofibrosis with myeloid metaplasia. *Br J Haematol* 113:763–771
- Tefferi A, Jiménez T, Gray LA, Mesa RA, Chen MG (2001b) Radiation therapy for symptomatic hepatomegaly in myelofibrosis with myeloid metaplasia. *Eur J Haematol* 66:37–42
- Tefferi A, Mesa RA, Hogan WJ, Shaw TA, Reyes GE, Allred JB, Ma CX, Dy GK, Wolanskyj AP, Litzow ML, Steensma DP, Call TG, McClure RF (2005) Lenalidomide (CC-5013) treatment for anemia associated with myelofibrosis with myeloid metaplasia (abstract). *Blood* 106:726a
- Terui T, Niitsu Y, Mahara K, Fugisaki Y, Urushizaki Y, Mogi Y, Kohgo Y, Watanabe N, Ogura M, Saito H (1990) The production of transforming growth factor- β in acute megakaryoblastic leukemia and its possible implications in myelofibrosis. *Blood* 75:1540–1548
- Thiele J, Kvasnicka HM (2002) CD34+ stem cells in chronic myeloproliferative disorders. *Histol Histopathol* 17:507–521
- Thiele J, Kvasnicka HM (2004) Prefibrotic chronic idiopathic myelofibrosis – a diagnostic enigma? *Acta Haematol* 111:155–159
- Thiele J, Kuemmel T, Sander C, Fischer R (1991) Ultrastructure of the bone marrow tissue in so-called primary (idiopathic) myelofibrosis-osteomyelosclerosis (agnogenic myeloid metaplasia). 1. Abnormalities of megakaryopoiesis and thrombocytes. *J Submicrosc Cytol Pathol* 23:93–107
- Thiele J, Kvasnicka HM, Boeltken B, Zankovich R, Diehl V, Fischer R (1999) Initial (prefibrotic) stages of idiopathic (primary) myelofibrosis (IMF) – a clinicopathological study. *Leukemia* 13:1741–1748
- Thiele J, Kvasnicka HM, Zankovich R, Diehl V (2001) Clinical and morphological criteria for the diagnosis of prefibrotic idiopathic (primary) myelofibrosis. *Ann Hematol* 80:160–165
- Thiele J, Imbert M, Pierre R, Vardiman JW, Brunning RD, Flandrin G (2001) Chronic idiopathic myelofibrosis. In: Jaffe ES, Harris NL, Stein H, Vardiman JW (eds) WHO Classification of tumours: tumours of haematopoietic and lymphoid tissues. IARC Press, Lyon, France, pp 35–38
- Thiele J, Kvasnicka HM, Schmitt-Gräff A, Diehl V (2003) Dynamics of fibrosis in chronic idiopathic myelofibrosis during therapy: a follow-up study on 309 patients. *Leuk Lymphoma* 44:549–553
- Thorns C, Rohrmoser B, Feller A, Horny HC (2002) Chronic cholecystitis and myeloid metaplasia. *Histopathology* 41:273–275
- Tsurumi S, Nakamura Y, Maki K, Omine M, Fujita K, Okamura T, Niho Y, Hashimoto S, Kanno K, Suzuki K, Hangaishi A, Ogawa S, Hirai H, Mitani K (2002) N-ras and p53 gene mutations in Japanese patients with myeloproliferative disorders. *Am J Hematol* 71:131–133
- Titus BR, Thiele J, Schaefer H, Kreipe H, Fischer R (1994) Ki-S1 and proliferating cell nuclear antigen expression of bone marrow macrophages. *Acta Haematol* 91:144–149
- Tsao M-S (1989) Hepatic sinusoidal fibrosis in agnogenic myeloid metaplasia. *Am J Clin Pathol* 91:302–305
- Tsakamoto N, Morita K, Maehara T, Okamoto K, Sakai H, Karasawa M, Naruse T Omine M (1994) Clonality in chronic myeloproliferative

- disorders defined by X-chromosome linked probes: demonstration of heterogeneity in lineage involvement. *Br J Haematol* 86:253–258
- Vannucchi AM, Bianchi L, Paoletti F, Di Giacomo V, Migliaccio G, Migliaccio AR (2004) Impaired GATA-1 expression and myelofibrosis in an animal model. *Pathol Biol (Paris)* 52:275–279
- Vannucchi AM, Bianchi L, Paoletti F, Pancrazzi A, Torre E, Nishikawa M, Zingariello M, Di Baldassarre A, Rana RA, Lorenzini R, Alfani E, Migliaccio G, Migliaccio AR (2005) A patho-biological pathway linking thrombopoietin, GATA-1 and TGF- β 1 in the development of myelofibrosis. *Blood* 105:3493–3501
- Varki A, Lottenberg R, Griffith R, Reinhard E (1983) The syndrome of idiopathic myelofibrosis: a clinicoathologic review with emphasis on the prognostic variables predicting survival. *Medicine (Baltimore)* 62:353–371
- Verstovsek S, Lawhorn K, Giles F et al (2003) PEG Intron therapy for patients with myeloproliferative diseases (MPD): interim analysis of phase II study (abstract). *Blood* 102:919a-920a
- Visani G, Finelli C, Castelli U et al (1990) Myelofibrosis with myeloid metaplasia: clinical and haematological parameters predicting survival in a series of 133 patients. *Br J Haematol* 75:4–9
- Visani G, Mele A, Malagola M et al (2003) Sequential combination of thalidomide and erythropoietin determines transfusion independence and disease control in idiopathic myelofibrosis previously insensitive to both drugs used as single agents. *Leukemia* 17:1669–1670
- Wang JC, Lang HD, Lichter S, Weinstein M, Bann P (1992) Cytogenetic studies of bone marrow fibroblasts cultured from patients with myelofibrosis and myeloid metaplasia. *Br J Haematol* 80:184–188
- Wang JC, Chen C, Lou LH, Mora M (1997) Blood thrombopoietin, IL-6 and IL-11 levels in patients with agnogenic myeloid metaplasia. *Leukemia* 11:1827–1832
- Wang JC, Chen C (1999) p16 gene deletions and point mutations in patients with agnogenic myeloid metaplasia (AMM). *Leukemia Res* 23:631–635
- Wang JC, Hemavathy K, Charles W, Zhang H, Dua PK, Novetsky AD, Chang T, Wong C, Jabara M (2004) Osteosclerosis in idiopathic myelofibrosis is related to the overproduction of osteoprotegerin (OPG). *Exp Hematol* 32:905–910
- Wanless IR, Peterson P, Das A, Boitnott JK, Moore GW, Bernier V (1990) Hepatic vascular disease and portal hypertension in polycythemia vera and agnogenic myeloid metaplasia: a clinicopathological study of 145 patients examined at autopsy. *Hepatology* 12:1166–1174
- Wernig G, Mercher T, Okabe R, Levine RL, Lee BH, Gilliland DG (2006) Expression of Jak2V617F causes a polycythemia vera-like disease with associated myelofibrosis in a murine bone marrow transplant model. *Blood* 107:4274–4281
- Williams ME, Innes DJ, Hutchison WT, Hesse CE (1985) Extramedullary hematopoiesis: a cause of severe generalised lymphadenopathy in agnogenic myeloid metaplasia. *Arch Intern Med* 145:1308–1309
- Wolf BC, Neiman RS (1987) Hypothesis: splenic filtration and the pathogenesis of extramedullary hematopoiesis in agnogenic myeloid metaplasia. *Hematol Pathol* 1:77–80
- Xu M, Bruno E, Chao J, Huang S, Finazzi G, Fruchtman SM, Popat U, Prchal JT, Barosi G, Hoffman R (2005) The constitutive mobilization of CD34+ cells into the peripheral blood in idiopathic myelofibrosis may be due to the action of a number of proteases. *Blood* 105:4508–4515
- Yan XQ, Lacey D, Hill D, Chen Y, Fletcher F, Hawley RG, McNiece IK (1996) A model of myelofibrosis and osteosclerosis in mice induced by overexpressing thrombopoietin (mpl ligand): reversal of disease by bone marrow transplantation. *Blood* 88:402–409
- Yotsumoto M, Ishida F, Ito T, Ueno M, Kitano K, Kiyosawa K (2003) Idiopathic myelofibrosis with massive ascites. *Intern Med* 42:525–528
- Zanke B, Squire J, Griesser H et al (1994) A hematopoietic protein tyrosine phosphatase (HePTP) gene that is amplified and overexpressed in myeloid malignancies maps to chromosome 1q32.1. *Leukemia* 8:236–244



<http://www.springer.com/978-3-540-34505-3>

Myeloproliferative Disorders
Melo, J.V.; Goldman, J. (Eds.)
2007, XI, 354 p., Hardcover
ISBN: 978-3-540-34505-3