

Polycythemia vera and the emperor's new clothes

In this issue of *Haematologica*, Passamonti and colleagues describe the clinical course of 70 polycythemia vera patients under the age of 50.¹ This is the largest series to date devoted to polycythemia vera patients in this age range and the one with the longest clinical follow-up. Najean *et al.* were the first to suggest that the clinical course of younger polycythemia vera patients might differ from that of their older counterparts.² In Najean's patients, initial disease manifestations with respect to blood counts and splenomegaly were more pronounced, the time between onset of symptoms and diagnosis shortened, and the incidence of significant thrombotic events at diagnosis, particularly intraabdominal venous thrombosis, higher. Remarkably, in spite of these burdens, life expectancy in this age group was longer (greater than 70% survival at 15 years). Two subsequent but smaller studies in patients under 45 years old also found a high incidence of significant thrombotic events at diagnosis as well as a continuing high incidence during follow-up,^{3,4} and in two series, the incidence of acute leukemia was increased,^{3,5} although in one, the sample size was small.³

In Passamonti's study, the incidence of thrombotic events was lower than in the previous studies, both at diagnosis and thereafter, the time between the onset of symptoms and diagnosis did not appear to be shortened, and the incidence of splenomegaly was also not as high, while life expectancy was similar (greater than 80% at 15 years). However, more importantly, Passamonti's patients had a high incidence of acute leukemia, confirming the earlier reports, while the incidence of myelofibrosis was much lower than in Najean's study. Passamonti's study is not only a useful addition to the few prior studies of polycythemia vera in younger patients but it also draws attention to issues beyond its immediate context and these issues form the basis of this editorial.

Is polycythemia vera a monolithic or a multifaceted disorder?

The most obvious issue is whether young polycythemia vera patients (less than 50 years old) constitute a unique population. I would propose, with one exception, that they do not, even though more recent data indicate that patients under 40 years of age constitute only 5% to 12% of patients with polycythemia vera^{2,6-8} and patients under 50 only 22% to 29%.⁸⁻¹⁰ However, it is easy to forget that until the second half of the twentieth century, younger patients were the norm not the exception.¹¹ The seminal case of Vaquez was 40 years old and had been symptomatic for 10 years.¹² Saundby and Russell's patient, one of the earliest recorded in the English literature, was 32 years old¹³ while Osler's index case was 44 and had been symptomatic for 5 years.¹⁴ The diminution in the size of the polycythemia vera patient population under the age of 50 has been a consequence of the increased longevity of the population in general and the well-documented

proclivity of the incidence of polycythemia vera to increase with age.^{15,16} Although, the natural history of disease manifestations appeared accelerated in some young patients,² this has not been a universal observation, and with the exception of an increased incidence of splenomegaly¹⁶ as well as intra-abdominal venous thrombosis in young women,^{2,18} the clinical manifestations of the disease are not different in this age group. The contention that the disease is not truly different in younger patients is bolstered not only by the rarity of the disease in childhood but also its generally milder manifestations in children as compared with any age group in adults.¹⁹ It is also noteworthy that patients with documented familial polycythemia vera usually do not manifest the disorder clinically until later in life.^{20,21} This suggests that a finite period is required for the necessary genetic changes to be acquired after the triggering insult before phenotypic expression of the disease can occur.

At the same time, leaving age considerations aside, it can be argued that polycythemia vera is not a monolithic disorder. This argument is bolstered by its varied initial clinical presentations – as idiopathic myelofibrosis,²² essential thrombocytosis²³ or idiopathic erythrocytosis¹⁵ – the varied tempo of disease progression in different patients, and their differing responses to treatment. Furthermore, although it was established many years ago that polycythemia vera, along with its companion myeloproliferative disorders, idiopathic myelofibrosis (IMF) and essential thrombocytosis (ET), is a clonal disorder arising in a multipotent hematopoietic progenitor cell,²³ more recent and extensive clonality studies suggest that in some patients, the disorder may be polyclonal²⁵ and occasionally, restricted to erythroid progenitor cells.²⁶ Similar observations have been made in ET,²⁷ including monoclonality restricted to platelets,²⁸ and while it has been suggested that clonal ET patients may have a higher incidence of thrombotic disease,^{27,29} the sample size was small and the issue remains unresolved.³⁰ Clonality assays, of course, are not without technical and interpretive problems,^{31,32} and these as well as incomplete clonal dominance, could give the appearance of polyclonality. Nevertheless, polyclonal malignancies are not unprecedented³³ and the development of clonally-derived hematopoiesis might only occur after a prolonged period of polyclonal hyperplasia, just as an adenomatous polyp precedes the development of a colonic carcinoma. The evolution of idiopathic erythrocytosis into polycythemia vera¹⁵ supports such a scenario. As a corollary, newer molecular markers such as impaired platelet Mpl expression³⁴ and granulocyte overexpression of PRV-1 mRNA³⁵ are most appropriately and usefully interpreted in this context,^{36,37} rather than as epiphenomena unworthy of attention.^{38,39}

Do we really understand the natural history of polycythemia vera?

Despite ten decades of scrutiny, the answer to this question is no. The conventional conception of polycythemia vera as a disease that passes through successive

phases — asymptomatic, erythrocytotic, quiescent, myelofibrosis with myeloid metaplasia and then possibly leukemia¹⁰ — was not derived in a manner that would satisfy present day standards of evidence⁴⁰ and has never been validated prospectively. Indeed, the variability in clinical presentation and complication rates in young patients evident in the studies by Passamonti and others, indicates that *one size does not fit all*. IMF should be the object lesson here. Once thought of as a disease of relentless progression with a life expectancy not greatly exceeding that of chronic myelogenous leukemia (CML),⁴¹ we now know that IMF is a heterogeneous illness and that the prognosis varies greatly, with survival being as long as 15 years in some patients.⁴² Remarkably and unfortunately, to date we have no such data for polycythemia vera patients because similar studies have not been performed in this more common illness although they are sorely needed.

Could polycythemia vera be hematology's Rosetta stone?

Although it is well established that polycythemia vera is a stem cell disorder, neither the specific stem cell involved nor its molecular defects have been identified. Defining either is unlikely to be a simple task, not only because we lack a clonal marker for polycythemia vera but also because the molecular defects are likely to be subtle in contrast to the gross disturbance of signal transduction that characterizes CML. Furthermore, even if a molecular lesion is identified, by analogy with CML,⁴³ it may well be a secondary defect, not the primary one. Malignant transformation is typically a multistep process and when one considers the slope of the incidence curve for polycythemia vera and the time before onset in most familial cases, it is likely that several steps are involved and, given the morphologically normal appearance of polycythemia vera hematopoietic cells, these steps are likely to be subtle ones. The ability of erythropoietin-independent erythroid progenitor cells to give rise to the erythropoietin-dependent progeny⁴⁴ as well as the differing time courses for the appearance of erythrocytosis, leukocytosis and thrombocytosis and the transitions between apparent ET,²³ IMF²² and polycythemia vera support these contentions.

At the same time, solving the riddle of polycythemia vera is likely to prove invaluable to the understanding of stem cell physiology in general. Indeed, at a time when interest in stem cells and their plasticity is at an all time high, it is difficult to understand why polycythemia vera is virtually ignored as a global model of a stem cell dysfunction. Both acute leukemia and polycythemia vera arise in a multipotent hematopoietic stem cell. However, while the leukemic blast cell can essentially be considered a *road kill* and its CD34⁺ ancestor the possessor of a repertoire of limited potential, the poly-

cythemia vera CD34⁺ cell possesses the potential for unparalleled and uncontrolled proliferation and plasticity as well as altered homing properties. Yet it produces phenotypically normal progeny. It thus represents a superb model system for studying biological processes whose mechanisms currently elude us. Polycythemia vera also provides an opportunity to study the influence of age on stem cell behavior, an influence suggested both phenotypically as well as by a related animal model⁴⁵ to be functionally stimulatory rather than inhibitory.

Are we treating polycythemia vera appropriately?

Remarkably, after ten decades of experience and an international co-operative group effort, the optimal treatment of polycythemia vera still eludes us. Although formation of the *Polycythemia Vera Study Group* (PVSG) was the correct and mandatory approach to the study of an uncommon disease of protean manifestations and chronicity, the effort was inevitably destined to fail. This was because well-documented blood volume physiology was ignored, unsubstantiated perceptions of the natural history of the disease were accepted uncritically, and, before any clinical trials, an unacceptable bias existed against phlebotomy,⁴⁶ the least toxic and most immediately effective therapy for the disease, while there was an unquestioned belief that thrombocytosis constituted a major risk for thrombosis in polycythemia vera⁴⁷ when it does not. The end result, well-documented in the major PVSG-01 clinical trial⁴⁷ and more recently in Passamonti's study as well as others,^{7,48} was treatment-related acute leukemia and bone marrow failure,^{49,50} suggesting that a young bone marrow is as susceptible to mutagens as an older one. Remarkably, none of Passamonti's patients received α -interferon. Since survival in Passamonti's patients was largely influenced by their exposure to marrow-damaging agents and not thrombosis, it should be abundantly clear that we have learned nothing from the PVSG experience. Indeed, a recent survey of the practice patterns of members of the American Society of Hematology in the post-PVSG era suggests that in general there is still no consensus as to the optimal approach to either the diagnosis or management of polycythemia vera.⁵¹

Conclusion: the emperor has no clothes

Passamonti's study, like similar prior studies, serves to remind us that we need to improve the management of polycythemia vera patients. Polycythemia may be an uncommon and chronic disorder but it is also a neoplasm and one for which we have no cure. Therefore, as for other hematologic malignancies for which there is no cure, its treatment should only be conducted in the context of well-controlled, prospective clinical trials. Given its low incident rate, this is only feasible through a co-operative group effort. The discovery of new molecular markers such as PRV-

1 and aberrant Mpl expression, the introduction of new therapeutic agents such as α -interferon⁵² and imatinib mesylate⁵³ and the growing evidence of the mutagenicity of drugs such as hydroxyurea⁵⁴ and pipobroman,⁵⁵ make such an approach not only attractive but also mandatory. Historically, from an organizational perspective, the PVSG experience provides a model for this as well as for avoiding its mistakes. What is needed now is the collective insight and will to do what is right for our patients, even though that means starting over again.

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FLT3 inhibition as tailored therapy for acute myeloid leukemia

The clinical success of the specific tyrosine kinase inhibitor, STI571^{1,2} (otherwise known as Glivec® or imatinib, Novartis Pharma), has fostered onco-hematologic research worldwide to develop new molecularly targeted forms of therapy. The target of imatinib is preferentially BCR-ABL, an intracellular oncogenic tyrosine kinase that shares several homologies with the class III receptor tyrosine kinase (RTK) family, whose members include the FLT3, KIT, FMS, and PDGF receptors.^{3,4} Most of these RTKs are implicated, either in mutated or wild-type conformations, in the constitutive activation and proliferation of human leukemias, especially acute myeloid leukemia (AML).^{3,5}

Particular interest has been aroused by the relatively high frequency of FLT3 receptor mutations found in AML. The FLT3 receptor has several structural domains, including 5 immunoglobulin-like domains in the extracellular regions, a juxtamembrane (JM) domain, 2 kinase domains (TK1 and TK2) separated by a kinase insert (KI) domain, and a C-ter-

minal domain in intracellular regions.⁴ Ligand binding to the RTK extracellular domain leads to receptor dimerization, stabilizes an open conformation of the catalytic domain (A-loop) for adenosine triphosphate (ATP) and substrate binding, and enables transphosphorylation of the A-loop. The subsequent phosphorylation of tyrosine residues accompanies RTK activation. Consequently, the FLT3 receptor leads to induction of fundamental intracellular signaling pathways, which in turn regulate both cell proliferation and apoptosis.³

The FLT3 receptor has been found to be frequently targeted in AML and somewhat less commonly in myelodysplastic syndromes (MDS) by two different types of genetic alteration.

Firstly, an internal tandem duplication (ITD) of the JM domain-coding sequence of the *FLT3* gene (FLT3/ITD) is found in 20% to 41% of adult and pediatric patients with *de novo* or secondary AML, as well as in about 3% of patients with MDS,⁶⁻¹⁰ as shown also by Moreno *et al.*¹¹ in this issue of *Haematologica*. These mutations constitutively activate the receptor, and are strongly associated with hyperleukocytosis, poor response to therapy and dismal prognosis. Among the distinctive forms of AML, higher frequencies of FLT3 alterations have been detected in acute promyelocytic leukemia (APL)¹² and in AMLs with apparently normal karyotype.⁶⁻¹¹ The reasons underlying these associations are currently unclear. When transplanted in murine hematopoietic progenitors, the mutant FLT3 receptor causes cellular transformation and produces a myeloproliferative syndrome, even though this does not by itself appear sufficient to cause acute leukemia.^{13,14} An additional length mutation affecting the tyrosine kinase domain in exon 20 has been recently described.¹⁵

Secondly, point mutations in the FLT3 receptor have been reported to occur in 3% to 8% of AML patients, mostly at a specific site in the gene (D835 and I836)¹⁶ (Figure 1). Although not apparently associated with either leukocytosis or worse prognosis, these point mutations result in similar deregulatory activity on the receptor and cause its constitutive activation and tend to worsen disease-free survival. Furthermore, the point mutations occur independently of FLT3/ITD. Taken together, these observations indicate that *FLT3* currently appears to be the most frequently mutated gene and constitutively activated receptor in AML.

The FLT3 receptor as a candidate target for tailored therapy in acute myeloid leukemia

Following the remarkable success of STI571, several researchers have pointed to the FLT3 receptor, or its signal transduction pathway,¹⁷ as a possible specific target for tailored therapy. Several tyrosine kinase inhibitors, which were not originally developed with FLT3 as the intended target, have been reported to inhibit the FLT3 receptor on AML cell lines or prima-