arm of chromosome 4 (start: 105,497,200 bp from pter; end; 106,825,780 bp from pter) (Figure 1A). Interestingly, the deleted region contained *TET2*, a gene recently found to be altered in many subtypes of myeloid malignancies[5-9] including 2 patients with RARS-T, of whom one showed a *TET2* missense and the other a frameshift mutation.[10] To further clarify the 4q24 deletion detected by SNP arrays, we performed fluorescence *in situ* hybridization (FISH). Twenty out of 100 analyzed interphase nuclei and three metaphases showed only one signal for the probe spanning the *TET2* gene in one patient (Figure 1B). Interphase FISH with the *TET2* probe was performed in 9 additional cases not analyzed by SNP arrays due to a lack of material. No additional case showing a deletion was detected. In addition to FISH, we performed *TET2* sequencing in 19/23 RARS-T. *TET2* mutations were detected in 5 out of 19 patients (26%), of which 3 out of 5 also presented mutated *JAK2*^{V617F}, whereas the remaining 2 out of 5 showed neither *JAK2*^{V617F} nor *MPL* nor *CBL* mutations. The 5 patients showed 6 individually different *TET2* mutations. Three were nonsense and two missense mutations. One patient displayed a frameshift mutation leading to a premature stop codon (Table 1). All mutations appeared to be heterozygous. The degree of homozygosity may, however, be underestimated due to a mixture of homozygous and healthy cells in the samples. As in other disease entities analyzed so far regarding *TET2* mutations, no mutation "hotspot" could be detected in our RARS-T patients. In summary, RARS-T patients show a high frequency of both *JAK2* and *TET2* mutations. Together with the less common *MPL* mutations described by others,[11,12] RARS-T presents a wide variety of mutations that overlap with the spectrum of mutations seen in MPN and other myeloid malignancies. Therefore, a combination of molecular markers including *JAK2* and *TET2* should be investigated to provide a more precise description of RARS-T as an independent entity.

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Key words: WHO classification, RARS-T, JAK2, TET2 mutations.

Acknowledgments: we thank all clinicians for sending RARS-T samples to our laboratory for diagnostic purposes and for providing clinical information.

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Citation: Flach J, Dicker F, Schnittger S, Kohlmann A, Haferlach T, and Haferlach C. Mutations of JAK2 and TET2, but not CBL are detectable in a high portion of patients with refractory anemia with ring sideroblasts and thrombocytosis. Haematologica 2010; 95:518-519. doi:10.3324/haematol.2009.013631

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Amyloidosis relapsing after autologous stem cell transplantation treated with bortezomib: normalization of detectable serum-free light chains and reversal of tissue damage with improved suitability for transplant

Systemic amyloidosis (AL) is a plasma cell dyscrasia in which the clone secretes free kappa (κ) or lambda(λ)-immunoglobulin light chains (FLCs).[1] These light chains do not fold into the proper tertiary conformation and form protein deposits, causing organ damage.[2] The most commonly affected organs are the heart, liver, kidney, gut, and peripheral nerves.[3] Standard treatment for patients with good performance status includes high-dose melphalan with autologous stem cell transplantation (ASCT).[1,4] Patients with organ dysfunction have increased transplant-related mortality.[5,6] Medications that treat AL without increasing the mortality of definitive treatment, currently ASCT, are sought. Bortezomib is a proteasome inhibitor that is effective in the treatment of plasma cell dyscrasias.[7] We utilized bortezomib to treat 2 patients with recurrent AL after initial ASCT. Both patients provided written informed consent according to the Helsinki Convention for their initial treatment, for ASCT, for bortezomib treatment of their relapsed disease, and for anonymous data collection.

Patient # 1, a 55-year old male, presented in April 2003 with severe congestive heart failure (CHF), renal failure, and bilateral pleural effusions. Congo red staining of myocardial biopsy indicated amyloid deposits (Figure 1). Serum λ-FLC level was elevated (Figure 2A). The patient received three monthly doses of melphalan 36 mg/m^2 after which his serum λ-FLC level was 3.38 mg/dL, his CHF resolved, with left ventricular ejection fraction increasing from 40% to 55%, and his renal failure improved (serum creatinine 1.2 mg/dL). The patient underwent ASCT with melphalan 140 mg/m^2 condition-

ing in November 2003; λ-FLC level decreased further to 1.92 mg/dL, and intraventricular septum wall thickness during diastoly (IVSD) decreased from 1.6 cm at diagnosis to 1.1 cm in August 2004. In January 2006, the patient relapsed; λ-FLC level increased to 5.34 mg/dL, IVSD increased to 1.5 cm, and peripheral edema with clinical CHF developed over one month. The patient received four cycles of bortezomib 1.3 mg/m^2 plus dexamethasone 20 mg (days 1, 4, 8 and 11, 21-day cycles) in February-April 2006 (Figure 2A), and experienced normalization of IVSD and diastolic dysfunction, λ-FLC level, and edema. The patient improved to such an extent (echocardiogram, N-terminal pro-brain natriuretic peptide, troponins, and CHF resolved) that he subsequently underwent an uneventful second ASCT with melphalan 200 mg/m^2 conditioning in June 2006, during which he experienced no cardiac arrhythmia, CHF, or significant orthostatic hypotension. As of March 2009, he remained in remission, with normal λ-FLC level and cardiac and renal function.

Patient # 2, a 58-year old female, presented in September 2004 with an elevated λ-FLC level of 62.4 mg/dL. Congo red staining of multiple gastrointestinal biopsies showed amyloid deposits. The patient was treated with three courses of melphalan 25 mg/m^2, during which λ-FLC level decreased to 8.9 mg/dL (Figure 2B). A colonoscopy in January 2005 showed that histological evidence of amyloid had resolved on biopsy samples. She underwent ASCT with melphalan 200 mg/m^2 conditioning in March 2005. At one month post-ASCT, her λ-FLC level had normalized (1.45 mg/dL), and remained normal for 18 months. By January 2007, λ-FLC level had increased to 9.38 mg/dL and gastrointestinal symptoms had recurred. The patient began five cycles of bortezomib plus dexamethasone (regimen as above) in January 2007 (Figure 2B). After three cycles, her λ-FLC level normalized, and her colonoscopy remained normalized at a follow-up examination in May 2007. The patient received two additional cycles beyond hematologic complete response. In May 2007, a colon biopsy showed extensive submucosal and vascular amyloid. In January 2008, λ-FLC level had increased to 3.38 mg/dL but she had no

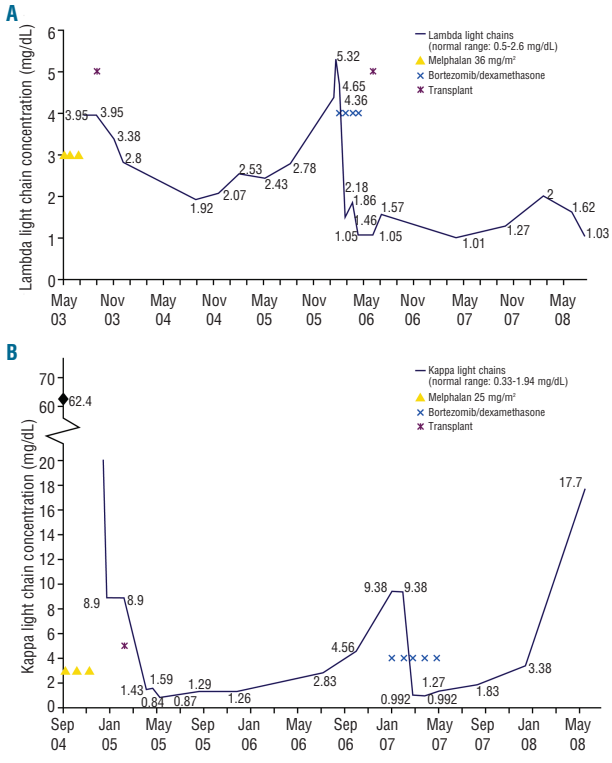

Figure 2. (A) Treatments of patient # 1 and response seen in the λ light chain level, and (B) treatments of patient # 2 and response seen in the κ light chain level.

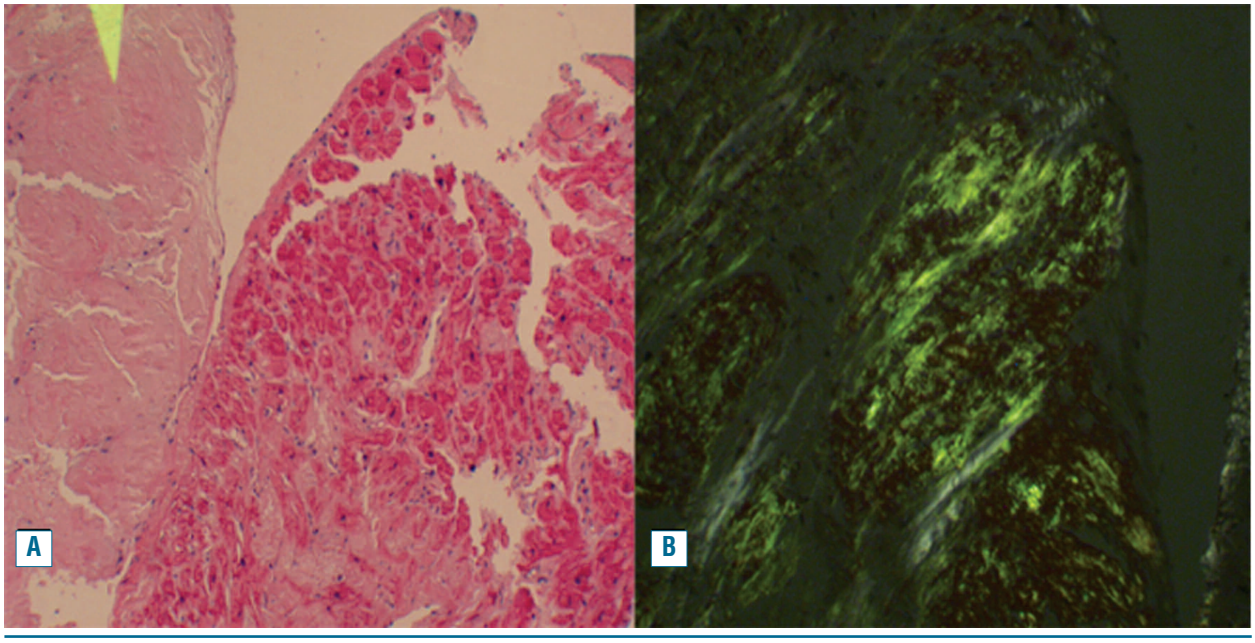

Figure 1. (A) Congo red stain of the cardiac biopsy from Patient 1 at the time of diagnosis. The biopsy is viewed under standard light, showing the pale eosinophilic amyloid fibrils within the cardiac muscle on the left. The disruption of the myocardium by the amyloid fibrils is seen best in the H&E. (B) Congo Red stain of the cardiac biopsy viewed under polarized light, showing the birefringent apple-green appearance of amyloid fibrils as they disrupt the cardiac muscle architecture.

gastrointestinal symptoms to indicate progression of tissue damage. Serial evaluation of λ-FLC level showed an increase to 17.7 mg/dL by June 2008. She has thus relapsed with increased λ-FLC level and gastrointestinal amyloid deposits on a recent biopsy, but has refused a second ASCT. Serum FLC data indicate she remained in molecular remission for more than 18 months after her last bortezomib treatment.

ASCT provides a substantial median survival in select AL patients,[8] but an over 25% mortality rate if even one organ has significant AL damage.[8,9] Our data suggest treatments that improve end-organ damage should reduce transplant-related mortality, ultimately allowing a higher percentage of patients to undergo ASCT with improved outcomes.[10] Bortezomib can reliably decrease serum FLC levels.[11] In our 2 patients, we documented reversal of AL organ damage with what appears to be promising disease-free survival. Furthermore, Patient 1, who received a second transplant, has remained in remission for three years. This suggests that second transplants may result in improved outcomes by significantly decreasing the AL disease burden. Treatment with bortezomib-based therapy may result in hematologic and organ responses that would enable patients with end-organ damage who would have otherwise been precluded from transplantation to undergo ASCT.

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Acknowledgments: the authors would like to thank Steve Hill and Jane Saunders of FireKite for editorial assistance in the preparation of this letter.

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Citation: Brunvand MW, Bitter M. Amyloidosis relapsing after autologous stem cell transplantation treated with bortezomib: normalization of detectable serum-free light chains and reversal of tissue damage with improved suitability for transplant. Haematologica. 2010; 95:519-521. doi:10.3324/haematol.2009.013102

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Successful eradication of acquired factor-VIII-inhibitor using single low-dose rituximab

It was with great interest that we read the recent recommendations regarding the treatment of acquired hemophilia A published in Haematologica[1] and followed the subsequent discussion.[2,3] The described treatment algorithm for this disease, which occurs at an incidence of 1.5/million/year,[4] stresses the importance of early eradication of the acquired fVIII-inhibitor. Using prednisolone alone or in combination with cyclophosphamide, this approach will be successful in 30-50% of affected individuals.[5] As Huth-Kühne pointed out, the CD20-antibody rituximab may be an attractive alternative in non-responders or patients with contraindications to steroids.[6] However, concerns have been raised because of rituximab side-effects including associations with progressive multifocal leukencephalopathy (PML).[7] Furthermore, as Mannucci *et al.*[2] emphasized, the use of this drug is hampered by its substantial costs. Restricting the administered dose to the lowest effective level, seems to be a reasonable strategy to minimize potential risks and costs of rituximab treatment.

We report a 66-year old male AHA-patient successfully treated with single low-dose rituximab. The patient presented with multiple hematomas and severe anemia after recovering from a respiratory infection. Prolonged activated partial-thromboplastin-time (aPTT) in combination with undetectable fVIII-activity due to an fVIII-inhibitor was observed. There was no clinical history of any prior hemorrhages nor a family history of bleeding. Clinical and imaging examinations could not reveal any pathological findings suggesting an idiopathic cause of AHA. Prednisolone (1 mg/kg) was started immediately

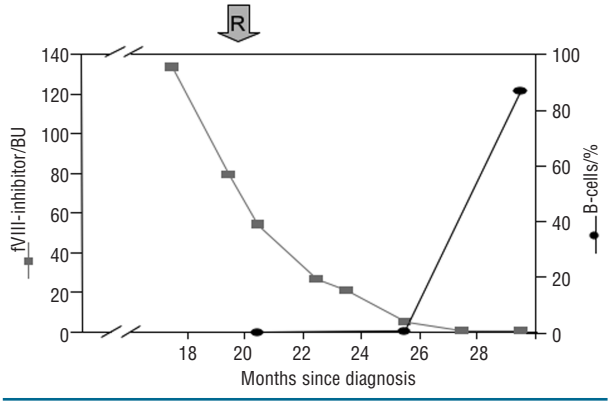

Figure 1. Clinical course overview. Time since diagnosis in months (x-axis). fVIII-inhibitor titer in Bethesda Units (BU) (left y-axis). B-cell proportion (%) (right y-axis). R: time of rituximab application.