rituximab, and TRAs that have strong evidence to support their use, tier 2 includes immunosuppressants (6-mercaptopurine, azathioprine, vincristine, and others), and danazol, for which there is less evidence of efficacy (often used in combination with tier 1 agents when monotherapy fails). Tier 3 strategies include plasmapheresis, stem cell transplantation, and interferon. Patient preference, comorbidities, and cost play a significant role in these decisions.

The study by Mahévas and colleagues reports the results of a multicenter analysis of 37 patients with multirefractory (to splenectomy, rituximab, and TRA) ITP compared with a matched group of 183 historical controls. Multirefractory patients had a significantly higher rate of secondary ITP (35.1%) than controls (8.7%). Even among patients with primary multirefractory ITP, half had biologic features of autoimmunity such as positive direct antiglobulin tests or antinuclear antibodies. Multirefractory patients also had higher rates of bleeding at onset (odds ratio, 3.54 [95% confidence interval, 1.12-11.23]; $P = .032$), low response rates to corticosteroids (68.6% vs 91.6%, $P = .002$), and a high mortality rate of 14%, only slightly lower than the 17.6% mortality rate among patients refractory to splenectomy reported by McMillan and Durette over a decade ago in the pre-TRA era.[3] However, an important finding in this study is that 7 of 10 patients with multirefractory ITP who were treated with a combination of TRA and 1 of several immunosuppressive therapies (5 mycophenolate, 1 cyclosporine, 2 cyclophosphamide, 1 azathioprine, and 1 hematopoietic stem cell transplantation) responded.

An array of diverse immunologic abnormalities involving T and B lymphocytes, dendritic cells, and plasma cells may contribute in different extents to the pathogenesis of ITP. Cines et al have suggested that ITP is not a specific disorder, but a "syndrome," and broadly categorized immune tolerance defects in this disorder as central defects that arise during early development, differentiation blocks with skewed B-cell subsets, and/or a loss of peripheral tolerance in the setting of immune stimulation.[6] Patients in whom acquired defects in peripheral tolerance checkpoints result in antibody-mediated platelet clearance and decreased megakaryopoiesis tend to respond well to conventional ITP therapy, frequently with durable responses, because the antigenic stimulus for autoimmunity may not be persistent. In contrast, patients with underlying autoimmune disorders and/or hematologic malignancies demonstrate a loss of central tolerance and differentiation blocks[6]; these patients have poor and short-lived responses to immune-directed therapy because additional cell types are involved in disease pathogenesis and the implicated lymphocyte repertoire is largely autoreactive and able to reconstitute rapidly after therapy (see figure). As demonstrated by this and other reports,[7,8] combination therapy directed at multiple arms of the immune response, along with TRAs, improves response rates. This is an important observation, though treatment of refractory ITP nevertheless remains reminiscent of the story of the "blind men and the elephant," in which a group of blind men touch different parts of an elephant, and when they compare notes find they are in complete disagreement. Unfortunately, personalized medicine has not yet been brought to bear on the management of ITP, though undoubtedly improved understanding of the mechanisms underlying autoreactivity could suggest targeted therapies with improved outcomes.

With new insights come new questions. Which immunosuppressive therapies are best combined with TRAs? Can we identify the predominant pathogenic mechanisms in individual patients to personalize therapy? Practically speaking, we suggest new definitions for refractory ITP in the era of TRAs that will keep everyone on the same page. We propose that ITP that fails or relapses after splenectomy be termed splenectomy-resistant ITP, whereas the term refractory ITP should be reserved for patients with persistent thrombocytopenia requiring treatment even after splenectomy, rituximab, and TRA.

Conflict-of-interest disclosure: The authors declare no competing financial interests. ■

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● ● ● CLINICAL TRIALS AND OBSERVATIONS

Comment on Fernandes et al, page 1555

Unchanneling cardiac iron in humans

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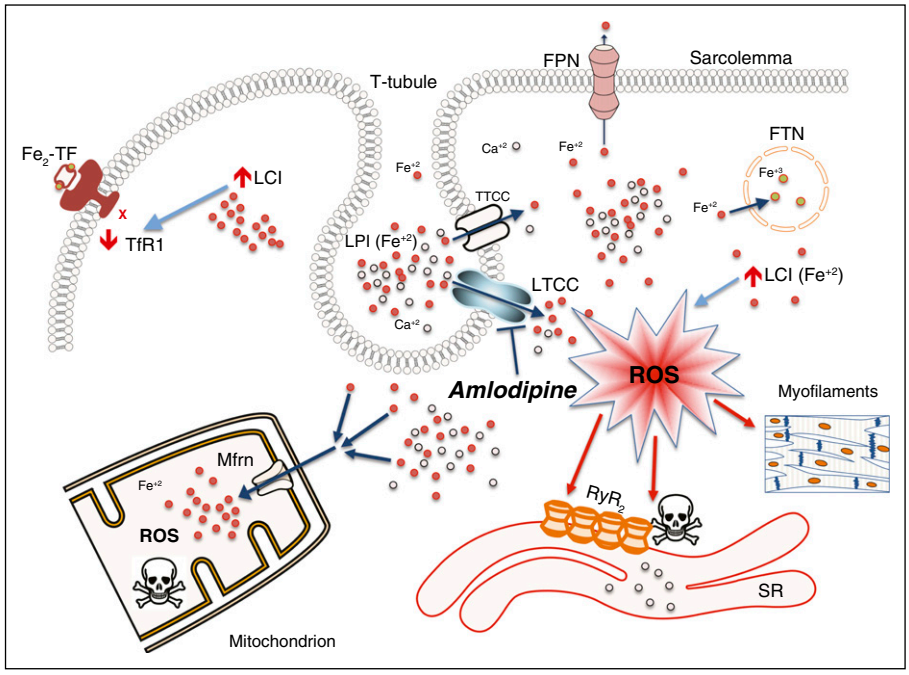
In this issue of *Blood*, Fernandes et al show that treatment of thalassemia patients with the calcium channel blocker amlodipine significantly reduces cardiac iron loading compared with placebo ($P < .02$).[1]

Thirty subjects with cardiac iron >0.59 mg/g dry weight heart tissue based on magnetic resonance imaging (MRI) were stratified to the iron "reduction" group and 29 with cardiac iron ≤0.59 mg/g cardiac tissue were placed in the "prevention" group. After 12 months, the



Iron transport through calcium channels in the heart. FPN, ferroportin; FTN, ferritin; LCI, labile cytosolic iron; LPI, labile plasma iron; LTCC, L-type calcium channel; Mfrn, mitoferrin; ROS, reactive oxygen species; RyR_2, ryanodine receptor; SR, sarcoplasmic reticulum; TF, transferrin; TfR1, transferrin receptor-1; TTCC, T-type calcium channel.

cardiac iron in the "reduction" group was 0.26 mg/g lower in the 15 subjects randomized to amlodipine plus standard iron chelation therapy compared with a 0.01 mg/g increase in the 15 subjects treated with placebo plus standard chelation ($P < .02$). Each of the subjects on amlodipine had a decrease in iron, and the overall average decrease was 21%. Amlodipine caused no change in liver iron concentration (LIC) or left ventricular ejection fraction.

So the question is, why is this small clinical trial of such pivotal importance in this day and age of massive multicenter prospective randomized studies?

The answer, in our opinion, is that this clinical study tells us that iron entry into the heart through L-type calcium channels (see figure), a mechanism that has been clearly demonstrated in vitro,[2-3] seems to be actually occurring in humans. As an added bonus, we have a possible new adjunctive treatment of iron cardiomyopathy.

The whole field of thalassemia and management of transfusional iron overload has made quantum advances in the past decade or 2. In the mid-70s, the median survival for thalassemia major was around 15 years of age, with death being secondary to cardiac complications of iron overload. By the early 2000s, mortality from iron cardiomyopathy had dropped by over 70%, thanks to effective chelators and the ability to routinely monitor organ iron concentration by MRI.[4] It is clear from serial monitoring by MRI that iron loading and unloading occurs at different rates in different tissues. The liver loads iron very quickly, and in fact the LIC is highly correlated with total body iron. However, there is a very poor correlation between the LIC and the pancreatic, pituitary, or cardiac iron concentrations, indicating that other factors control iron trafficking in these organs.[5] The explanation lies in the very elegant iron regulatory biochemistry that has been elucidated over the past 2 decades. Normally, iron circulates bound to TF and enters cells by receptor-mediated endocytosis via the TF receptor, TfR1. When LCI (Fe^{+2}) increases, TfR1 transcription decreases, preventing cellular iron overload. However, when total iron body increases dramatically, as is the case in patients on chronic transfusion, the TF-binding ability is rapidly exceeded and circulating non-TF bound iron (NTBI) appears in the plasma. NTBI rises considerably when the TF saturation reaches ~60%, and a highly reactive Fe^{+2} subspecies of NTBI called LPI increases concomitantly. This is when problems begin. Humans do not normally have circulating Fe^{+2}. LPI can then enter cells through ion transporters that are normally designed to carry divalent cations like zinc and calcium. For the most part, these ion transporters are not regulated by intracellular iron concentration and iron loading proceeds, even though cytosolic iron levels may be very high. Various levels of expression of divalent metal transporters (DMT1, ZIP14) and of the iron exporter, ferroportin have been demonstrated in the liver, pancreas, erythroid precursors, and the pituitary of animal models, and the proportion of transporters that have their transcription regulated by iron is organ specific. For example, there are few iron response elements (IRE)-containing DMT1 messenger RNA splice variants in the pancreas, whereas all DMT1 splice variants in the brain contain an IRE. Thus, the pancreas loads Fe^{+2} in the face of high intracellular iron, whereas the brain does not.[5] The heart also contains L-type and T-type calcium channels,[3] both being quite capable of transporting Fe^{+2} into the heart, which of course brings us to the L-type calcium channel blocker, amlodipine (see figure).

This influx of Fe^{+2} into the heart via calcium channels has several consequences.[3,6] Fe^{+2} competes with the entry of Ca^{+2} into the heart via calcium channels. This can have a significant effect on cardiac excitation contraction coupling. The elevated Fe^{+2} in the cardiomyocytes increases the production of intracellular ferritin, the protein that binds toxic Fe^{+2} and converts it to the nonreactive Fe^{+3}. LCI in the cytosol is highly toxic and produces reactive oxygen species that poison the ryanodine receptors in the sarcoplasmic reticulum that are critical for myocyte function.[3] Lastly, Fe^{+2} is transported into myocardial mitochondria leading to mitochondrial iron overload and dysfunction that can further affect myocardial contractility. Interestingly, severe mitochondrial iron loading can cause relative depletion of cytosolic iron, resulting in a positive feedback loop that increases iron flux into the myocyte.[7] This probably explains the clinical observation that cardiac function and cardiac iron levels remain stable for years, but once loading starts, cardiac iron loading and dysfunction progress very rapidly over a few months, often leading to death.

The problem is that all of this seemingly coherent and elegant iron trafficking story, including the idea that the L-type calcium channel blocker amlodipine, as well as other calcium channel blockers can modulate cardiac iron trafficking, is derived from in vitro studies in cell culture and from knockout mice. Certainly, serial measurements showing differential rates of loading and unloading during chelation in the liver, endocrine organs, and heart in humans are consistent with the elegant biochemistry worked out in the laboratory.[5] However, some hand waving is required to explain this organ-specific iron trafficking in humans, because mice are not humans. Fernandes et al have shown that the effect of calcium channel blockade amlodipine on cardiac iron loading predicted in animals also resulted in reduced cardiac iron in humans. As the authors point out, more clinical studies are needed, and certainly biochemical studies need to continue because all calcium channel blockers do not have the same effect in vitro,[3] but at least the "channels" for more progress on both clinical and biochemical fronts are now open.

Conflict-of-interest disclosure: The author declares no competing financial interests. ■

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Comment on Chen et al, page 1562

BV for HL: can the responses last?

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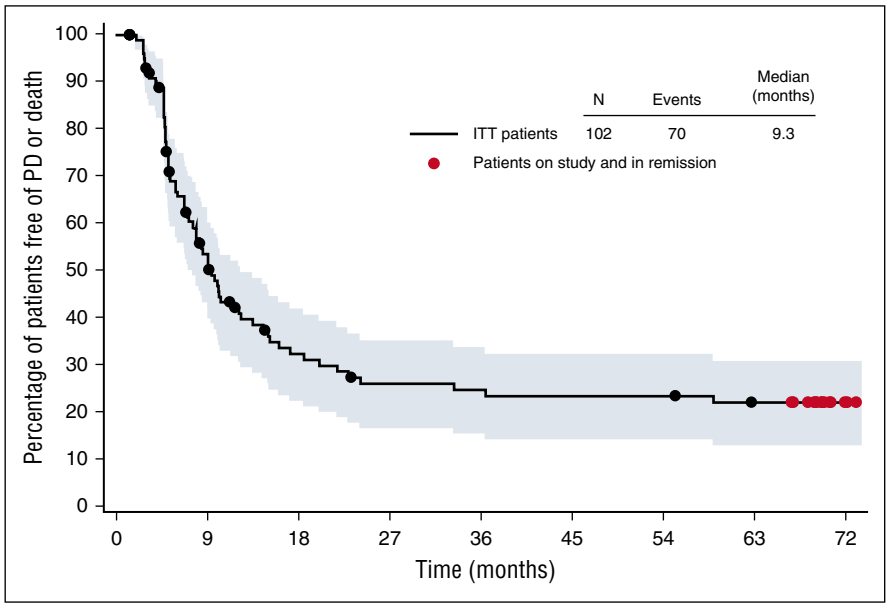
In this issue of *Blood*, Chen et al report the long-term follow up from a cohort of relapsed/refractory (R/R) Hodgkin lymphoma (HL) patients, who received single-agent brentuximab vedotin (BV) in a phase 2 pivotal trial, revealing a 5-year overall survival (OS) and 5-year progression-free survival (PFS) of 41% and 22%, respectively (see figure).[1]

The original report from this trial demonstrated an overall response rate of 75% and a complete response (CR) rate of 34%, leading to accelerated US Food and Drug Administration (FDA) approval for patients with R/R HL after hematopoietic autologous stem cell transplantation (auto-SCT) or failure of at least 2 prior therapies.[2] BV, a novel antibody drug conjugate, has been described as a "game changer" in HL. Is BV living up to the hype? Of the 102 patients enrolled, 13 remain in continuous complete remission >5 years from registration. Of the 13 patients, 9 achieved this result having received only BV, whereas 4 received BV followed by hematopoietic allogeneic SCT (allo-SCT). Although 9% durability for BV alone may not seem like a noteworthy outcome, recall that these are generally young patients who had received 2 prior attempts at curative therapy (frontline therapy and auto-SCT). Given that the median age of the cohort was only 31, any degree of remission durability is meaningful. Long-term follow up also affords an opportunity to examine long-term toxicity. In the original publication on this cohort, 42% of patients experienced peripheral neuropathy (PN), of which 8% was grade 3.[2] PN, even grades 1 to 2, is often a source of daily aggravation and frustration for patients. In this updated report, we learn that 88% of patients experienced either resolution (73%) or improvement (14%) in symptoms.

Do these results signify a significant therapeutic advance? Yes. Could similar results have been obtained with already available agents and strategies? Maybe, but patients likely would have endured more toxicity



PFS in 102 HL patients treated with BV. See Figure 1B in the article by Chen et al that begins on page 1562.

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Unchanneling cardiac iron in humans

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