the power of adenovirus vectors for clinical gene transfer via the systemic circulation. Even in children, one finds cross-reacting anti-adenovirus Abs that can opsonize or neutralize vectors. Add to this the fact that erythrocytes have CAR on their membranes,[7,8] platelets bind some adenovirus types, and that the fenestra size in human liver may be near the limit to allow the approximately 90-nm adenovirus particle to enter. We see that we still have work to do before one can master the fate of adenovirus vectors in the circulation. Nonetheless, the field now has one less hurdle to cross.

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

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● ● ● CLINICAL TRIALS

Comment on Witt et al, page 952

Avoiding "sticker" shock

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In this issue of *Blood*, Witt and colleagues show that it is safe to alter the frequency of laboratory monitoring from every 4 weeks to every 8 weeks in stable patients on long-term anticoagulant therapy.[1]

Long-term administration of a coumarin drug is currently the only option for chronic oral anticoagulation therapy in the United States. Coumarins are effective and inexpensive, but close monitoring and frequent dose adjustments are usually required to achieve the desired antithrombotic effect and avoid bleeding. Monitoring requires venipuncture, performing a prothrombin time, and calculating the International Normalized Ratio (INR). Studies have established that an INR of 2 to 3 (or 2.5 to 3.5 in patients with mechanical heart valves) best balances safety and efficacy. The task of the clinician is to determine the dose of drug that consistently produces an INR in the therapeutic range. Current recommendations call for testing to begin 2 to 3 days after therapy is initiated. Once the INR is therapeutic, the monitoring frequency is decreased to 2 to 3 times weekly for 1 to 2 weeks, and if the INR remains in the therapeutic range, eventually to once every 4 weeks for as long as the patient is on the drug.[2] Patients whose INRs remain in the therapeutic range on repeated testing are considered to have achieved stable anticoagulation.

Can the frequency of monitoring be decreased in patients on stable anticoagulation? This question has been addressed by Witt et al in this issue of *Blood*. They conducted an observational study of more than 6000 patients followed by a centralized clinical pharmacy anticoagulation service. Forty-one percent had all INR values within the therapeutic reference interval for a continuous 6-month interval (stable patients), and the remainder (59%) served as comparator patients. Patients had at least 1 INR determination every 8 weeks. In the comparator group, only 47% of INR values were in the therapeutic range, and this group had a significantly higher rate of bleeding (2.8% vs 0.8%; $P < .001$). Stable patients were less likely to have a goal INR ≥ 3.0 (OR = 0.48, 95% CI 0.38-0.61). Other predictors of a stable INR were age older than 70, and absence of diabetes, heart failure, or other chronic disease. Thus, the authors have identified and characterized patients potentially requiring less frequent anticoagulant monitoring. To confirm that such patients can be safely managed with less testing, they suggest a prospective trial of stably anticoagulated patients in whom the interval between INR determinations is progressively increased to 8 or even 12 weeks.

Table 1. Explanations to patients taking coumarins (5)

1.	Why treatment is necessary
2.	How the drug works
3.	Why monitoring is needed
4.	Why it is important to take the drug at a fixed time daily
5.	How alcohol may affect anticoagulation
6.	How dietary changes affect therapy
7.	How drug-drug interactions might affect treatment
8.	Why clinic personnel should be notified of changes in medications
9.	What precautions should be taken to avoid bleeding
10.	How to recognize signs and symptoms of bleeding

While such a suggestion is reasonable, the dismal track record of coumarins with respect to bleeding must be recognized. Despite careful monitoring of anticoagulant intensity, the risk of major hemorrhage in several large clinical trials has ranged from 0.3% to 0.5% per year as compared with controls,[3] and during long-term anticoagulation, 1.1% per patient-year.[4] Some of the hemorrhages might have occurred because of unpredictable events such as intercurrent illnesses or trauma. Changes in patient medications and use of nonprescription drugs and alcohol might have precipitated other bleeding events. Perhaps the best approach to limit these complications is a well-informed patient. In this regard, the 10 key issues for patient explanation (see table) are as valid today as they were when published 25 years ago by Errichetti et al.[5] These issues should be continually reinforced while patients are taking these drugs. Well-educated patients who have achieved stable anticoagulation are candidates for less frequent monitoring and fewer needle sticks.

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LYMPHOID NEOPLASIA

Comment on Hideshima et al, page 1046

Bortezomib paradigm shift in myeloma

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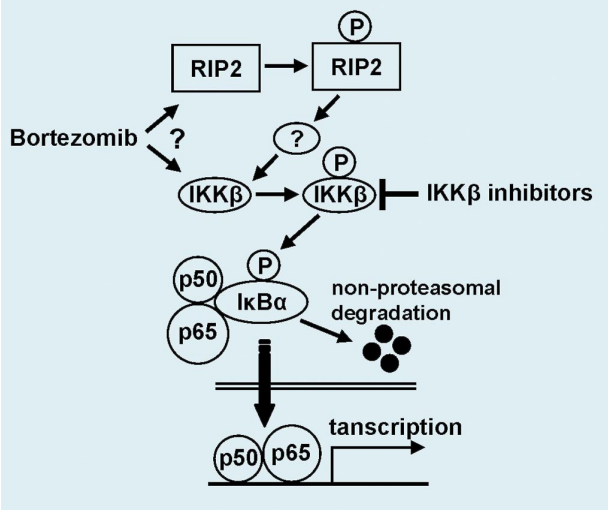
Bortezomib's unprecedented antitumor activity in myeloma has long been attributed to NF-κB inhibition. A new study from the group that put the drug on the map directly challenges this assumption.

Bortezomib (Velcade, formerly PS-341) is a peptide boronate inhibitor of the proteasome that was developed for cancer therapy by Julian Adams, Peter Elliott, and their colleagues.[1] At the time, this was considered a very bold move, since loss of proteasome function was thought to be incompatible with viability in normal cells. However, using a novel assay that directly measures 20S proteasome activity, they demonstrated that rodents and primates tolerated doses of bortezomib resulting in up to 80% proteasome inhibition without obvious toxicity. They also used this assay to guide dose escalation in cancer patients in phase 1 clinical trials in solid and hematologic malignancies.[1] Although single-agent activity was modest in most tumors, Anderson's group led a phase 2 clinical trial that showed that bortezomib was active in relapsed refractory multiple myeloma (MM),[2] leading to FDA approval of the drug for the treatment of MM in 2003.

Although it was assumed from the start that bortezomib would have diverse effects on cancer cell biology, the most common mechanism attributed to its antitumor actions was inhibition of the inflammation-associated transcription factor, NF-κB. Work performed by several groups implicated NF-κB activation in the maintenance of cancer cell survival,[3] and other studies demonstrated that conventional cancer therapies also commonly activated NF-κB, undermining their therapeutic potential.[4] Preclinical studies and clinical trials confirmed that bortezomib down-regulated tumor cell expression of known NF-κB transcriptional targets (IL-6, etc), consistent with the hypothesis. Furthermore, a more recent high-profile study demonstrated that the subset of primary MMs that possesses activating mutations within the so-called non-canonical NF-κB pathway was especially sensitive to bortezomib,[5] adding further support to the concept.

A cautionary note was introduced by work performed several years ago, showing that NF-κB inhibition did not fully account for bortezomib's cytotoxic effects in MM cells.[6] Nonetheless, it is still generally accepted that bortezomib is a potent and general NF-κB inhibitor and that these effects contribute to cytotoxicity. In this issue of *Blood*, the new work by Hideshima et al turns this idea on its head.[7] Using human MM cell lines and primary tumor specimens, they now show that bortezomib actually activates 2 upstream NF-kB–activating kinases (RIP2 and IKKβ), promotes down-regulation of NF-κB's inhibitor (IκBα), and increases NF-κB DNA binding in vitro (see figure). Another structurally unrelated proteasome inhibitor (lactacystin) induces the same effects, strongly suggesting that NF-κB activation is an "on-target" effect of the drug. Furthermore, they present evidence that bortezomib also fails to block NF-κB in vivo, determined by measuring nuclear localization of NF-κB's RelA/p65 subunit.[7] Exposure of cells to bortezomib in the presence of a selective IKK antagonist (MLN120B) blocks NF-κB activation and promotes cell death. Together, these results provide compelling evidence that NF-κB inhibition is probably irrelevant to the effects of bortezomib in MM cells.

Why did it take so long for this long-held assumption to be overturned? One reason is that bortezomib's documented NF-κB inhibitory effects were almost always measured in cells exposed to cytokines that are known to promote proteasome-dependent degradation of IκBα (ie, TNFα), while investigators largely ignored bortezomib's lack of inhibitory effects on basal NF-κB activity. In addition, the possibility that nonproteasomal degradation of IκBα might contribute to NF-κB activation was not explored. So, if NF-κB inhibition is not involved in bortezomib's cytotoxic effects, what other mechanisms might explain its unique potency in MM cells? One attractive explanation (and the simplest) is that cell death results from protein build-up and aggregation,[8,9] as is the case in neurodegenerative diseases.[10] In this model, the high levels of immunoglobulin production and ER-Golgi



Possible mechanism whereby bortezomib triggers canonical NF-κB activation. Bortezomib either directly or indirectly (via RIP2) activates IKKβ, which subsequently phosphorylates IκBα, an inhibitor of p50/p65. After nonproteasomal degradation of IκBα, p50/p65 translocates to nucleus. IKKβ inhibitors block down-regulation of IκBα and NF-κB activity as well as enhance bortezomib-induced cytotoxicity. See the complete figure in the article beginning on page 1046.