IRF7$^{-/-}$ mice. It is unclear whether these discrepancies are due to differences between the species. However, the observed abrogation of immune dysfunction induced by TLR7 ligands in MyD88$^{-/-}$ and TLR7$^{-/-}$ knockout mice strongly suggests the involvement of TLR7 and its downstream pathways.

Taken together, these recent studies are consistent with a model in which chronic stimulation of the innate immune system by TLR ligands, either encoded by HIV-1 or found in the circulation after microbial translocation from the gut, result in the chronic production of proinflammatory cytokines driving generalized immune activation and disease progression in HIV-1–infected individuals. Similar to hepatitis C virus infection, another chronic persistent viral infection in which genetic polymorphisms in TLR7 have been associated with differences in liver fibrosis,[7] recent studies have associated polymorphisms in TLRs with differential HIV-1 disease outcome,[8] further supporting this model. At this point, studies in humans are needed to examine this model and its significance for HIV-1 pathogenesis. The recent developments of TLR antagonists that can block TLR signaling make such studies possible. The reduction of chronic immune activation via TLR stimulation might represent an attractive potential target to reduce immunopathology in chronic HIV-1 infection.

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

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● ● ● RED CELLS & IRON

Comment on Jiao et al, page 462

Ironing out complementary medicine

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At least some of the beneficial effects attributed to the complementary medicine agent curcumin may result from its iron chelating properties.

Turmeric, a yellow powder derived from the root of the flowering plant *Curcuma longa*, is both a dye and an aromatic spice frequently used in Asian and Indian cooking.[1] It also has a long history of use in Eastern traditional medicine for a wide variety of disorders. The active agent in turmeric is the polyphenol curcumin, which has been shown to have antineoplastic activity in vitro and chemopreventive effects in animal models of induced cancer. When combined with its favorable toxicity profile, these features have helped curcumin make the transition "from the kitchen to the clinic."[2]

Curcumin has been reported to have a wide variety of cellular and molecular effects, including inhibition of NFκB, COX2 activation, Akt inhibition, and a variety of redox effects, both pro-oxidant and antioxidant. In this issue of *Blood*, Jiao and colleagues demonstrate that curcumin can act as an iron chelator in vivo.

In a previous report, the same group had noted in vitro reduction of cellular ferritin by curcumin and suggested that curcumin had moderate iron chelating activity.[3] In this present study, the authors first establish baseline hematologic and iron parameters for mice fed diets that ranged from minimal iron (5 mg/kg added iron) to excessive iron (1000 mg/kg added iron). The addition of curcumin induced a state of overt iron deficiency anemia in mice on the marginal iron diet and produced evidence of iron mobilization in mice at all dietary iron levels, as indicated by declines in liver ferritin and increases in liver transferrin receptor-1 and iron regulatory protein activity. Curcumin decreased hepcidin expression. The biologic relevance of these changes was confirmed by demonstration of consequent increases in cellular ferroportin protein.

Although this study is not the first to propose iron chelation as an effect of curcumin,[4] it is the first to thoroughly demonstrate iron chelation in vivo and to confirm this by demonstrating predicted changes in the iron regulatory system. The study has limitations: for valid technical reasons, the indicators of iron mobilization studied differed between mice on the high iron diet and the marginal iron diet, and wide variation of hepcidin expression in the low iron diet control mice precluded demonstration of a statistically significant effect of curcumin on hepcidin in vivo. The hepcidin/ferroportin results described above were therefore demonstrated using the HepG2 cell line. The important question of whether the variety of cellular effects reported for curcumin are consequences of its iron chelating activity or are independent of this activity was outside the scope of this report.

A search of the website www.clinicaltrials.gov in October 2008 showed 23 therapeutic or chemopreventive trials using curcumin in a wide variety of diseases. The 3 most frequently studied were colorectal cancer, pancreatic cancer, and Alzheimer disease. Other diseases under study included multiple myeloma, myelodysplastic syndrome, rheumatoid arthritis, and psoriasis. The report by Jiao et al leads to a number of important questions relevant to the clinical use of curcumin. Does the tolerability of large doses of curcumin offset its relatively modest chelating activity, making it a viable therapeutic option for clinical chelation? Does its chelating effect impose limitations on its use as adjunctive or chemopreventive therapy in patients with marginal iron stores? Marginal or absent iron stores are common in patients with colon cancer,[5] the disease in which curcumin is most commonly studied at present. A similar concern would potentially apply to its use in women with limited iron stores due to menstrual or gestational

The root of iron chelation? Reprinted with permission from the California Department of Food and Agriculture.

blood loss. For patients with hypoferremia due to the anemia of chronic disease, would curcumin exacerbate anemia, or would its effects on hepcidin alter the effects of hepcidin on other aspects of erythropoiesis in a favorable direction?[6] Like much good research, the current report answers some questions while giving rise to new ones.

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

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● ● ● TRANSPLANTATION

Comment on Paczesny et al, page 273

Graft-versus-host disease: proteomics comes of age

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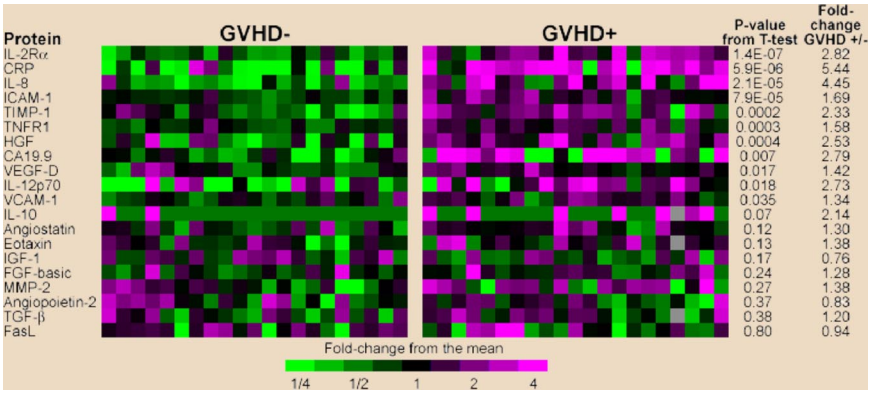
In this issue of *Blood*, Paczesny and colleagues describe a set of 4 serum proteins that may help to confirm the diagnosis of acute GVHD and seem to be associated with worse survival independently of GVHD grade.



ELISA heat map of discovery set samples. Gray indicates that the sample was not assayed for that protein. Levels of PSA-ACT, IL-17 and IL-1β were not detectable and therefore do not appear in the figure. Please see the complete figure in the article beginning on page 273.

Acute graft-versus-host disease (GVHD), the main complication of allogeneic hematopoietic cell transplantation, is mainly diagnosed clinically and can be confirmed by biopsy of 1 of the 3 target organs (skin, gastrointestinal tract, or liver). The severity of acute GVHD is graded clinically from I to IV, with increased mortality rates in severe GVHD (grades II-IV).

Although multiple blood proteins have been described as potential biomarkers in previous smaller studies, no single protein or panel has emerged with sufficient specificity and sensitivity to enter clinical use. More recently, mass spectrometric profiling[1-3] of urine and serum were reported to demonstrate the presence of spectral patterns associated with GVHD, but these approaches do not identify specific proteins.

In this issue of *Blood*, Paczesny et al aimed to isolate candidate proteins using high-throughput assays on a large number of patient samples, and to determine their significance with respect to patient outcome. They screened plasma with antibody microarrays for 120 proteins in a discovery set of 42 transplantation patients that revealed 8 potential biomarkers for diagnostic of GVHD. Using enzyme-linked immunosorbent assays (ELISA), they then measured the levels of these biomarkers in samples from more than 400 transplantation patients divided into training and validation sets. Statistical analysis of these 8 proteins determined a panel of 4 proteins (IL-2–receptor–alpha, TNF-receptor-1, IL-8, and hepatocyte growth factor) that discriminated patients with and without GVHD. In patients with GVHD, Cox regression analysis revealed that the biomarker panel predicted survival independently of GVHD severity. This study is a major achievement in the field and brings proteomics nearly from the bench to the bedside. The authors have to be congratulated for the huge amount of work they have done and the refined analysis of their results.

However, as always in good science, this study raises more questions than it answers: As mentioned, other authors have used mass spectrometric profiling (these approaches do not identify specific proteins) while Paczesny et al used a predefined set of 120 proteins. In the first case, we are faced with lovely diagrams[1-3] without knowing if they constitute proteins actually involved in inflammatory processes or allogeneic reactions. In the second