

symptoms other than chest pain are of value in identifying patients with suspected coronary artery disease who should undergo functional testing.

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Selective Adhesion-Molecule Therapy and Inflammatory Bowel Disease — A Tale of Janus?

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Although our understanding of the pathogenesis of the chief forms of inflammatory bowel disease, Crohn's disease, and ulcerative colitis remains incomplete, progress is being made in identifying essential components.¹ The presence of large numbers of varied leukocytes within affected tissue where they are normally sparse makes it axiomatic that active disease is dependent on the recruitment of these cell populations. Recruitment is now known to proceed through a stereotypical series of steps that depend on selective adhesion molecules (SAMs). These include cell-surface integrins, heterodimers formed by various combinations of α and β subunits. Integrins with an α_4 chain appear to play an especially important role in the intestine.² $\alpha_4\beta_1$ Integrin (a combination also known as very late antigen 4 [VLA-4]) is present on most leukocytes but not neutrophils and effects binding to vascular-cell adhesion molecule 1 on endothelium and dendritic cells. $\alpha_4\beta_1$ Integrin can also mediate binding to components of the extracellular matrix. In contrast, $\alpha_4\beta_7$ integrin is expressed on subpopulations of lymphocytes, natural killer cells, and monocytes and selectively targets them to so-called gut-associated lymphoid tissue. Thus, in the

latter guise, α_4 integrin mediates tissue-specific transport of cells to the intestine.

Circumstantial and direct experimental evidence has suggested that α_4 integrins are important in the recruitment and activation of cells in inflammatory bowel disease. Tissues affected by inflammatory bowel disease have increased levels of α_4 integrins and their ligands.³ Moreover, a disease remarkably similar to ulcerative colitis spontaneously develops in cotton-top tamarins, and administration of a monoclonal antibody against α_4 integrin led to the resolution of colitis in these animals.⁴ A subsequent study also showed that treatment with an antibody specific for $\alpha_4\beta_7$ integrin was beneficial in the cotton-top tamarin model.⁵

Over the past several years, insights into the mechanisms of recruitment have prompted efforts to develop agents to address unmet medical needs of patients with inflammatory bowel disease. A number of these agents are currently being evaluated, but the study that is farthest along is that of natalizumab, a humanized IgG4-class monoclonal antibody directed against α_4 integrins that has already been approved for the treatment of patients with multiple sclerosis. Two early studies suggested

that natalizumab had an effect in patients with Crohn's disease, but the results fell short of statistical significance.^{6,7} These studies served as the foundation for larger, tandem studies — the Efficacy of Natalizumab as Active Crohn's Therapy (ENACT-1) trial and the Evaluation of Natalizumab as Continuous Therapy (ENACT-2) trial — whose results are reported in this issue of the *Journal*.⁸ The ENACT-1 study randomly assigned patients with moderate-to-severe Crohn's disease to receive either natalizumab or a placebo in a fixed-dose regimen of three infusions given over an eight-week period. Patients who reached the primary end point of a response (defined by a reduction in the Crohn's Disease Activity Index score of at least 70 points) at 10 weeks in the ENACT-1 study were randomly reassigned in the ENACT-2 study to receive either natalizumab or placebo every 4 weeks through week 56.

In the ENACT-1 study, a response was not significantly more common among those receiving natalizumab than among those receiving placebo. An especially high rate of response to placebo (49 percent) in this trial may have made it difficult to observe a treatment effect. The high frequency and durability of the placebo response in the ENACT-2 study arouse concern that the results may have been partially confounded by the enrollment of some patients with little or no active disease. This possibility is consistent with the findings in a post hoc analysis of a lower rate of placebo response among patients with elevated levels of C-reactive protein, those receiving concomitant immunosuppressive therapy, and those with a previous response to therapy against tumor necrosis factor α (TNF- α).

In contrast to the ambiguous results of the ENACT-1 study, among patients who had a response to natalizumab in the first study, the rates of sustained response during the ENACT-2 study were significantly higher among those who continued to receive natalizumab rather than being given placebo (61 percent vs. 28 percent, $P < 0.001$). The median time to the loss of response was also much longer among those receiving the antibody. The higher frequency of smokers among those receiving placebo in the ENACT-2 study, as compared with those receiving natalizumab (26 percent vs. 16 percent), may have confounded the results, since smoking has been associated with increased activity of Crohn's disease. Overall, the positive findings in

the ENACT-2 study suggest that the mechanisms sustaining disease activity and those sustaining mucosal homeostasis may be distinct.

For natalizumab, the first anti-SAM therapy, to add a new dimension to the treatment of inflammatory bowel disease, it must have an acceptable risk of adverse events. The ENACT studies found a relatively low rate of immunogenicity or loss of responsiveness. More germane is the apparent unintended consequence of immunodeficiency. Indeed, virtually every agent currently used in the treatment of inflammatory bowel disease creates an iatrogenic form of immunodeficiency and a risk of opportunistic infection. No doubt, the deleterious effects on immune defenses represent the other Janus-like face of the mechanism that confers therapeutic benefit. Some opportunistic infections seem to be selectively associated with individual treatments, presumably illuminating mechanisms important in specific immune defense; for example, the reactivation of latent tuberculosis after the administration of anti-TNF- α therapy represents a clinical experiment that illuminated the key role played by TNF- α in the control of mycobacteria and other intracellular pathogens.

Recent reports suggest that studies of natalizumab therapy may similarly delineate mechanisms necessary for immune control of the nearly ubiquitous JC virus, a human polyomavirus. Progressive multifocal leukoencephalopathy has developed in at least three patients receiving natalizumab (one with Crohn's disease who received the agent during an open-label extension study after early discontinuation from the ENACT-2 study), two of whom died.⁹⁻¹¹ This development has prompted the manufacturer to halt sales of the antibody for approved uses.

Up to 80 percent of the adult population is infected with the JC virus. The virus is likely encountered in childhood and then persists in a latent form within the epithelium of the gastrointestinal tract and kidney throughout life. Activation of the latent virus and subsequent dissemination have been causally linked to the development of progressive multifocal leukoencephalopathy.¹²⁻¹⁴ The presence and dissemination of JC-virus-specific cytolytic T cells have been reported in patients with human immunodeficiency virus infection or AIDS, suggesting that these T cells may play an important role in the control of latent infection.¹⁵ The cluster of cases among patients receiving natalizumab provides

evidence that α_4 integrin-dependent pathways are an essential part of host defenses that control latent JC virus. Identifying those pathways remains an important challenge that may ultimately define the limits of the use of natalizumab and the overall potential for α_4 integrin-based therapy with antibodies against SAMs. It will be important to determine which integrin ($\alpha_4\beta_1$, $\alpha_4\beta_7$, or both) is necessary for host control of JC virus. Resolving this mechanistic question is clearly important to an assessment of the potential risks or advantages of even more specific anti-SAM therapy with antibody against $\alpha_4\beta_7$ integrin — an approach that also shows promise in the treatment of inflammatory bowel disease.¹⁶ Finally, it should be noted that JC virus has also been potentially linked to gastrointestinal tract cancers, particularly those of the colon; reactivation of the virus may contribute to the chromosomal-instability pathway.^{17,18} If correct, this would raise at least a hypothetical concern about the use of this agent in patients with inflammatory bowel disease.

Having found through unfortunate clinical experience that antibody against α_4 integrins may inhibit mechanisms necessary for the control of latent JC virus infection and the prevention of progressive multifocal leukoencephalopathy, could this approach nonetheless be a useful addition to treatment options for inflammatory bowel disease? Two observations may help frame an appropriate answer to this question. First, it is possible that prospective magnetic resonance imaging (MRI) can identify progressive multifocal leukoencephalopathy in asymptomatic patients. Second, other agents with clinically significant risks of inducing potentially life-threatening immunodeficiency have clear value to patients with inflammatory bowel disease and their physicians. For example, one large series documented a rate of death of 1 percent among those receiving infliximab for more than a year.¹⁹ So, as in any clinical challenge, the question is whether the potential benefit is worth the risk. Our understanding of the benefits of natalizumab in patients with Crohn's disease is too incomplete to offer a definitive answer. Neither is it appropriate to dismiss the value of the agent because of the risk of a complication — which although serious, might be monitored — when so many patients' needs are not adequately met by available therapies. Reason would suggest that natalizumab and subsequent anti-SAM therapies should be targeted to patients who have frequent and debilitating

recurrences of inflammatory bowel disease and should be given in conjunction with close surveillance by MRI for the earliest signs of progressive multifocal leukoencephalopathy. In the meantime, efforts should be focused not only on defining the true therapeutic efficacy of this and related agents but also on gaining a more precise understanding of the mechanisms of action and risk of treatment with antibody against α_4 integrins. In time, pinpointing the cause of the major forms of inflammatory bowel disease should diminish our need to treat what appears to be a naturally occurring form of immunopathy with man-made forms of selective immunodeficiency.

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Metabolic Disorders in the Center of Genetic Medicine

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Genetics is the new frontier of medicine. Hardly an issue of any leading medical journal is published without one or more articles on a genetic disease or a topic closely related to genetics. A recent series of articles in the *Journal* was devoted to the importance of genetics in medical practice.¹ The immediate impetus for such a series was, as is widely known, the sequencing of the human genome and the promise that that effort would lead to better diagnosis and treatment of disease.² However, the knowledge that many metabolic disorders are genetic has long been seen as evidence, with clear implications for medical practice, that inherited genetic traits and propensities play a large part in disease.

At the center of knowledge about inherited metabolic diseases has been phenylketonuria, a disorder of phenylalanine metabolism, and newborn screening and dietary treatment have virtually eliminated mental retardation in patients with this disorder. From the understanding of the genetic basis of phenylketonuria has come the realization that there is a tangible benefit from diagnosing inherited disorders.³ Phenylketonuria also stimulated genetic research and federal funding of this research. One of the first-cloned genes involved in genetic disease was that for phenylalanine hydroxylase, the enzyme that is defective in phenylketonuria.⁴ The excitement of understanding phenylketonuria led to frequent metabolic testing of children with clinical disease that might be metabolic, especially those with developmental delay or other neurologic symptoms. Such strategies resulted in the discovery of many additional metabolic disorders. The need to understand the disorders motivated investigations that began to elucidate not only the metabolic defects but also areas of human biochemistry and pathology. Thus, as Garrod realized in the early 1900s as a result of his studies of alkaptonuria that led to the concept of the inborn error of metabolism, the importance of these disorders transcends their rar-

ity; they can provide paths to greater knowledge about normal human biology and to deeper understanding of common problems.⁵

Homocystinuria provides a particularly striking example of this importance. This disorder is a well-delineated inborn error of methionine metabolism that causes ectopia lentis, skeletal abnormalities, mental retardation, and thromboembolism. Its cardinal biochemical feature is an increased level of homocysteine. A few years after homocystinuria was reported, Harvey Mudd and I and our colleagues studied an infant who not only had increased levels of homocysteine but also had other biochemical abnormalities that are not found in homocystinuria, including an increased level of methylmalonic acid. Eventually, we recognized that the infant had a new disorder in which vitamin B₁₂ was not metabolized to the cobalamin coenzymes necessary to stimulate remethylation of homocysteine to methionine and to convert methylmalonic acid to succinic acid.⁶

On autopsy of this infant, McCully found vascular occlusions that resembled those seen in patients with homocystinuria; he suggested that the increase in the level of homocysteine was the common etiologic factor of the occlusions.⁷ Studies stimulated by these findings have now shown that almost 10 percent of the general population is homozygous for a thermolabile variant of methylenetetrahydrofolate reductase, an enzyme that is also responsible for remethylation. These people usually have hyperhomocysteinemia, which is considered a risk factor for cardiovascular disease.⁸ The reduced activity of methylenetetrahydrofolate reductase can be restored by administration of large amounts of folate, which lowers the level of homocysteine. These observations provide the basis for the current measurement of plasma homocysteine during health examinations and constitute the major reasons for the recommendation of an increase in folate intake and of a daily supplement of folic ac-