The science of leukocyte adhesion began with the observations of children with what we now call leukocyte adhesion molecule 1 (LAD 1) disease. Affected children have recurrent infections, delayed umbilical cord separation, leukocytosis, and a paucity of leukocytes in infected tissue (1). Molecular elucidation of the deficiency in CD11/CD18 expression in these children coupled with the recognition and characterization of other leukocyte adhesion molecules (LAMs) such as integrins, immunoglobulin supergene family, selectin and addressin families has led to the burgeoning field of leukocyte adhesion biology. Leukocyte adhesion molecules have been demonstrated to mediate a myriad of cellular processes, ranging from "inside-out signaling" to regulation of transcription and apoptosis (2). Nevertheless, whenever studies have been performed to evaluate the role of LAMs in mediating inflammatory diseases, most of the tissue effects have been associated with and attributed to leukocyte migration: either the presence or absence of a particular cell type after blockade, or genetically deleting a leukocyte adhesion pathway. This emphasis on leukocyte recruitment, stemming from the initial observations of impaired leukocyte recruitment in children with LAD 1, has to some degree impeded the translational studies on the role of LAMs in inflammatory diseases, with clinically relevant research lagging sorely behind the rapid developments in basic adhesion biology.

Asthma is a disease of increased incidence and severity associated with industrialization. With the abundance of inflammatory cells in bronchiolar lavage and airway tissue biopsies associated with airway hyperreactivity (AHR) to inhaled cholinergic and other bronchoconstrictive agonists, testing the utility of LAM blockade in models of asthma is a logical step. The VLA-4 molecule is particularly attractive to block because it is expressed on eosinophils and lymphocytes, which have been implicated in the late allergic airway response of asthma. Simplistically thinking, blocking leukocytes from entering the lung from the circulation in sensitized individuals could abrogate airway inflammation and AHR. Surprisingly, VLA-4 or combined CD11a and CD11b antibodies given to sensitized rats block the early airway response (EAR) to allergen provocation testing, which is an event that is dependent on mast cell activation and degranulation (3). Expression of adhesion molecules on mast cell surface presumably accounts for this finding and suggests a mechanism of action related to interference with cell activation rather than migration.

In previous studies, VLA-4 antibody given intravenously blocked the late airway response (LAR), suggesting the importance of cell migration in the phenomenon. However, this treatment did not have much influence on the migration of leukocytes into airway tissues or airway lumen (3). Similar effects were seen with combined CD11a and CD11b blockade. The discrepancy between the inhibition of the LAR by anti–VLA-4 and insignificant effects on leukocyte migration was confirmed in sheep and suggestive evidence of an effect on eosinophil activation was obtained (4). In this issue of *The Journal*, Henderson et al. have helped to clarify this issue by examining the effects of anti–VLA-4 on measurements of airway responsiveness to methacholine, leukocyte migration, cytokine production, and mucous secretion in a mouse model of "asthma" (5). An intranasal administration of VLA-4 antibody abrogated AHR and TH2 cytokine release to ovalbumin challenge without blocking circulating VLA-4 in the blood, while intraperitoneal VLA-4 antibody decreased airway eosinophilia but had no effect on AHR. By colocalizing the VLA-4 binding of intranasally administered antibody to a CD11c-positive intrapulmonary leukocyte (possibly a dendritic cell or activated macrophage), they suggest that the effects of anti–VLA-4 are on resident cells and are not related to leukocyte migration into lungs after challenge from AHR. Furthermore, the evidence argues against the eosinophil as the causative agent of hyperresponsiveness. A similar finding has been reported by Milne and Piper for the guinea pig (6) and by Laberge et al. for the rat (7). The inhibition of the T cell cytokines associated with allergic inflammation (TH-2 cytokines, IL-4, and IL-5) by intranasal antibody is also an interesting observation and is consistent with inhibition of T cell activation. It is conceivable that interference with antigen presentation may account for the reduction in T cell cytokines. The link between effects on T cell function and AHR has not been made in this study, however.

The effectiveness of intranasal and thus intraairway administration is very appealing for another reason. LAM blockade has the potential to cause systemic susceptibility to infection, thus organ-specific blockade is preferred. However, systemic VLA-4 administration may still be effective despite the finding that intraperitoneal administration was not effective in blocking AHR in the current study (5). This is presumably a reflection of the administered dose which may have been too low to block intrapulmonary VLA-4 receptors.

How does one reconcile these observations with the multitude of studies that have demonstrated that preventing leukocyte migration to inflamed organs is associated with decreased tissue injury? Clearly, LAMs have numerous functions. Thus, while leukocyte migration is easy to detect and quantify, leukocyte function in tissue is complex and the modification of function may escape detection. In addition, the type of immune injury is important, such that ischemic reperfusion injury, classically associated with neutrophil migration, probably engages LAMs in a different manner than other inflammatory processes, such as allograft rejection. Organ specificity is also likely to be important, as tissue-specific vascular beds and nuances in blood flow and matrix are likely to make conclusions from one organ inapplicable to another without direct demonstration. Unlike muscle or heart (where LAM blockade is tissue protective in ischemic reperfusion injury, with clear effects on abrogating leukocyte recruitment), the interpretation of responses of the kidney is full of inconsistencies. CD11/CD18 and ICAM-1 blockade are usually protective in experimental renal ischemia–reperfusion injury, but systemic neutropenia and leukopenia, as well as selectin blockade, are often not (reviewed in reference 8). As in the lung, LAM blockade in kid-

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ney is likely abrogating a key step in tissue dysfunction independent of leukocyte migration. There are many possible cellular pathways, including activation, apoptosis, and ion transport, which may be crucially altered and deserve attention.

Several questions arise regarding LAMs and asthma. What are the cellular effector functions that VLA-4 and perhaps other LAMs are mediating that are so vital to AHR? It is important to remember that adhesion molecules may participate in other aspects of the pathogenesis of allergic airway inflammation. Several LAMs participate in T cell responses to sensitization as costimulatory signals. As it is not clinically feasible in most situations to treat just before antigen challenge, one must consider the potential for the induction of T cell tolerance by interfering with these costimulatory signals. There are other aspects of airway function in asthma that merit attention. For example, what is the interplay between VLA-4 and the smooth muscle abnormalities that characterize asthma? How do the LAMs, particularly ICAM-1, play a role in infection by airway viruses (e.g., rhinovirus, respiratory syncytial virus) that often precede an asthma attack? What cellular assays can be used in tissue to best assess leukocyte function within tissue? It is likely that effective treatment for some inflammatory diseases will be based on leukocyte adhesion molecules. To make this feasible, it will be necessary to accept revelations from a disease rather than preconceptions of how these molecules work.

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References

1. Arnaout, M.A. 1990. Leukocyte adhesion molecule deficiency: its structural basis, pathophysiology and implications for modulating the inflammatory response. *Immunol Rev.* 114:145–180.

2. Hynes, R.O. 1992. Integrins: versatility, modulation, and signaling in cell adhesion. *Cell.* 69:11–25.

3. Rabb, H., R. Olivenstein, T.B. Issekutz, P.M. Renzi, and J.G. Martin. 1994. The role of the leukocyte adhesion molecule VLA-4, LFA-1 and Mac-1 in allergic airway responses in the rat. *Am. J. Respir. Crit. Care Med.* 49:1186–1191.

4. Abraham, W.M., M.W. Sielczak, A. Ahmed, A. Cortes, I.T. Lauredo, J. Kim, B. Pepinsky, C.D. Benjamin, D.R. Leone, R.R. Lobb, and P.F. Weller. 1994. Alpha$_4$-integrins mediate antigen-induced late bronchial responses and prolonged airway hyperresponsiveness in sheep. *J. Clin. Invest.* 93:775–787.

5. Henderson, W.R., E.Y. Chi, R.K. Albert, S.J. Chu, W.J.E. Lamm, Y. Rochon, M. Jonas, P.E. Christie, and J.M. Harlan. 1997. Blockade of CD49d (α_4 integrin) on intrapulmonary but not circulating leukocytes inhibits airway inflammation and hyperresponsiveness in a mouse model of asthma. *J. Clin. Invest.* 100:3083–3092.

6. Milne, A.A.Y., and P.J. Piper. 1994. The effects of two anti-CD18 antibodies on antigen-induced airway hyperresponsiveness and leukocyte accumulation in the guinea pig. *Am. J. Respir. Cell Mol. Biol.* 11:337–343.

7. Laberge, S., H. Rabb, T.B. Issekutz, and J.G. Martin. 1995. Role of VLA-4 and LFA-1 in allergen-induced airway hyperresponsiveness and lung inflammation in the rat. *Am. J. Respir. Crit. Care Med.* 151:822–829.

8. Rabb, H., Y.M. O'Meara, P. Maderna, P. Coleman, and H. Brady. 1997. Leukocytes, adhesion molecules and ischemic acute renal failure. *Kidney Int.* 51:1463–1468.