MMP-9) activity and thereby limit aberrant elastin degradation, and/or by limiting excessive TGF- β signaling, perhaps by preventing TGF- β activation, or TGF- β release from the extracellular matrix, and/or by limiting pulmonary inflammation. However, none of these possibilities have been experimentally confirmed. In addition, the authors have not considered any direct effects of elastin on the innate and adaptive immune system, recently reviewed by Sallenave (13). A mechanistic explanation for their observations will go a long way to deepen our understanding of the mechanistic basis of BPD, and should be addressed in future studies.

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The Lung Macrophage: A Jack of All Trades

It is well known that resident macrophages play an essential role in the innate immune response in the lung. However, emerging evidence suggests that macrophages play not one, but many different roles in the development and resolution of acute lung injury and lung fibrosis. In this issue of the *Journal* (pp. 547 and 569), two groups of investigators examined the role of macrophages in the development and resolution of lung injury and fibrosis in chronic and acute murine models (1, 2). These investigators found evidence for distinct populations of macrophages in the lung that undergo dramatic changes in both number and phenotype during the development and resolution of lung injury. Both groups of investigators found that manipulation of the macrophage pool in the lungs of mice alters the severity and persistence of lung injury and fibrosis, suggesting that macrophages play a causal role in the pathophysiology of these disorders.

There are two broad subsets of macrophages in human and mouse lung: alveolar macrophages, which line the surface of alveoli; and interstitial macrophages, which are localized in the narrow space between the alveolar epithelium and vascular endothelium (3). Landsman and Jung suggested that interstitial macrophages originate from monocytes and may serve as an obligatory intermediate between blood monocytes and alveolar macrophages (4). Using several surface markers, macrophages

can be further characterized as exhibiting an M1 phenotype (classically activated macrophages) or an M2 phenotype (alternatively activated macrophages) (5). M1 macrophages are generally associated with a Th1 immune response, the production of reactive oxygen intermediates, and robust bacterial killing. M2 macrophages are associated with Th2 immune response, immune response to helminthes, tissue healing, collagen production, and fibrosis. These characteristics suggest that M1 macrophages are the first to respond to lung injury or infection and are later replaced by M2 macrophages that contribute to tissue fibrosis or repair.

Gibbons and colleagues used two complementary techniques to deplete macrophages and their precursors—monocytes—in mice following the induction of lung fibrosis by the intratracheal administration of bleomycin or intratracheal infection with an adenovirus encoding an active form of TGF- β 1 (2). The deletion of macrophages at a point in time when fibrosis was progressing in the bleomycin model significantly reduced the severity of the resulting fibrosis. In contrast, depleting macrophages during the resolution of bleomycin-induced fibrosis delayed its resolution. The investigators observed increased staining of lung macrophages with two markers of alternatively activated macrophages: Ym1 in the mouse and CD206 in patients with idiopathic pulmonary fibrosis (IPF). These findings are consistent with a recent report by Sun and coworkers (6), who found an increased number of alternatively activated macrophages in a model of lung fibrosis induced by overexpression of IL-10, and with those of Pechkovsky and colleagues (7) and Mathai and coworkers (8), who observed increased expression of CD206—another marker of alternatively

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activated macrophages—in patients with IPF and systemic sclerosis, respectively. Collectively, these findings suggest that alternatively activated macrophages play an important role in the development and resolution of lung fibrosis after injury and raise the intriguing possibility that persistent M2 activity might contribute to the failure to resolve fibrosis in patients with IPF.

Because the systemic administration of liposomal clodronate decreased both inflammatory monocytes and alternatively activated macrophages in the lung, Gibbons and colleagues hypothesized that the alternatively activated macrophages are derived from circulating monocytes. To test this hypothesis, they adoptively transferred Ly6C^{hi} bone marrow–derived monocyte progenitor cells into bleomycin-treated mice during the progressive phase of pulmonary fibrosis. This led to a significant exacerbation of pulmonary fibrosis and an increased number of alternatively activated lung macrophages in the lung. Surprisingly, the increased numbers of alternatively activated macrophages were host derived and not from the donor Ly6C^{hi} population. Additional studies that employ sorted blood monocytes, control injections of Ly6C^{low} monocytes, and use additional markers for alternatively activated macrophages will be required to more clearly identify the macrophage pool from which M2 macrophages in the fibrotic lung are derived.

In another article, Janssen and colleagues (1) studied the fate of resident and recruited lung macrophages during acute lung injury. The investigators used syngeneic bone marrow transplantation into lethally irradiated mice to show that the steady-state turnover of alveolar macrophages in the lung is surprisingly low: 3 months after bone marrow transfer, approximately 70% of alveolar macrophages were of recipient origin, and even 8 months after bone marrow transfer more than 70% of alveolar macrophages remained host derived. These results are similar to those of Maus and colleagues (9), who reported that 3 and 12 months after bone marrow transplantation approximately 85% and 60% of alveolar macrophages, respectively, were still host derived. They then examined the role played by resident and recruited macrophages during acute lung injury induced by the intratracheal administration of LPS or infection with the H1N1 influenza A virus. They found that the resident alveolar macrophages pool remained static throughout the duration of the lung injury and the expansion of the lung macrophage pool was mediated by an influx of the donor-derived monocytes from the peripheral blood, followed by their differentiation into tissue macrophages. Fate mapping experiments suggested that the recruited macrophages appeared to be cleared by apoptosis and phagocytosis by neighboring macrophages, and not by migration into the lymphatic nodes. The investigators observed that resident macrophages expressed lower levels of Fas compared with the recruited macrophages, perhaps explaining their persistence. To test this hypothesis, they transferred bone marrow from Fas mutant (lpr) mice and administered small molecule inhibitors of caspase-8 (which is activated upon Fas ligation) systemically to mice. Mice receiving Fas-mutant bone marrow and mice treated with a caspase-8 inhibitor displayed a delayed resolution of acute lung injury. In contrast, mice treated with a Fas-activating antibody recruited fewer macrophages to the injured lung. These results suggest that apoptosis of the recruited macrophages may be required for the resolution of acute lung inflammation. Other groups of investigators have reported that the ingestion of apoptotic bodies by macrophages yields an antiinflammatory-activated macrophage phenotype (10). It is tempting to speculate that these macrophages contribute to the resolution of lung injury or that macrophages that elude apoptotic clearance might represent a source of alternatively activated macrophages that might contribute to the development of lung fibrosis. The tools

described by both Gibbons and coworkers and Janssen and colleagues should help us to address these questions.

Together, these studies reveal that the development and resolution of lung injury is accompanied by dramatic changes in not only the numbers but also the types of macrophages in the lung. These studies demonstrate a causal role for macrophages in determining the severity of both the acute and fibrotic phases of lung injury. Furthermore, both of these studies suggest that clearance of recruited macrophages is important for the resolution of lung injury and normal repair, opening a novel area of investigation to identify new therapies to treat patients with acute lung injury and lung fibrosis.

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