Asbestos-Induced Alveolar Injury*
Evidence for Macrophage-Derived PDGF as a Mediator of the Fibrogenic Response

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The cellular and biochemical mechanisms that mediate injury and repair in any fibrotic lung disease largely are obscure. We have used asbestos exposure in rats and mice to provide a model of rapidly progressing interstitial fibrosis. Our previous studies have shown that inhaled fibers are translocated through the type I alveolar epithelium and attract lung macrophages through complement-dependent mechanisms. Within 24 h after exposure, nonciliated epithelial cells of the terminal bronchioles, type II epithelial cells, and interstitial fibroblasts of the bronchiolar-alveolar duct junctions exhibit up to 20-fold increases in incorporation of thymidine into DNA. In addition, endothelial and smooth muscle cells of small vessels in this anatomic region also show dramatic proliferative changes. Inasmuch as lung macrophages are a prominent anatomic feature of the developing fibrotic lesions and produce a remarkable armamentarium of cytokines, we are testing the postulate that these cells are the central mediators of the fibrogenic process. Platelet-derived growth factor (PDGF) accounts for a significant amount of mesenchymal cell growth-promoting activity found in macrophage-conditioned medium. Thus, in ongoing studies we are measuring the amount of PDGF secreted by macrophages after inhalation of chrysotile asbestos and a nonfibrogenic particle, carboxyl iron.

Studies
Alveolar macrophages were collected by lavage 1 wk after a 3-h exposure to either iron (50 mg/m²), asbestos (10 mg/m²), or room air. Macrophages (37 x 10⁶) from 3 to 4 rats were cultured in serum-free medium for 24 h and PDGF was quantified in the macrophage-conditioned medium by enzyme immunoassay. For comparison, macrophages lavaged from unexposed animals were activated in vitro by adding particulate suspensions to plastic-adherent macrophages for 24 h. Macrophage-conditioned medium was concentrated 100-fold, acidified to 1 M acetic acid for 1 h, and fractionated on a Superose 12 FPLC gel filtration column. Neutralized fractions were tested for PDGF by using an enzyme immunoassay. The data represent mean ± SEM of 3 separate experiments.

Discussion
These data show that inhalation of asbestos and iron upregulates the secretion of PDGF by lung macrophages and that the activity of PDGF is modulated by α-M. We postulate that asbestos may cause pulmonary fibrosis by persisting in the lung and causing a long-term activation of the alveolar and interstitial lung macrophage populations to secrete cytokines that mediate cell growth, chemotaxis, and extracellular matrix production. Because PDGF is a potent

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References
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Figure 1. Secretion of a platelet-derived growth factor homologue by alveolar macrophages (M0s) following in vitro or in vivo activation via the phagocytosis of chrysotile asbestos or carboxyl iron. Macrophages activated in vitro were lavaged from animals 1 wk post exposure and cultured for 24 h. Macrophages lavaged from unexposed animals were activated in vitro by adding particulate suspensions to plastic-adherent macrophages for 24 h. Macrophage-conditioned medium was concentrated 100-fold, acidified to 1 M acetic acid for 1 h, and fractionated on a Superose 12 FPLC gel filtration column. Neutralized fractions were tested for PDGF by using an enzyme immunoassay. The data represent mean ± SEM of 3 separate experiments.
stimulator of fibroblast growth and chemotaxis, ongoing studies are designed to address the mechanisms of PDGF biologic activity and if it is central to the progression of pulmonary fibrosis.

**REFERENCES**


**Analysis of Local mRNA Expression for Extracellular Matrix Proteins and Growth Factors Using in situ Hybridization in Fibroproliferative Lung Disorders**

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Fibroblast activation with subsequent deposition of extracellular matrix (ECM) proteins occurs in the reparative processes following a wide variety of lung injuries. Morphologic studies employing immunohistochemical localization demonstrate enhanced procollagen type I and fibronectin deposition in the fibroproliferative lesions observed in idiopathic pulmonary fibrosis (IPF) and bronchiolitis oblit-

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**RESULTS**

Tissue from patients with IPF contained two distinct lesions generally thought to represent early and later stages in the fibrotic process. Inflammatory exudates comprised predominantly of macrophages were observed in otherwise normal-appearing alveolar air spaces. These lesions contained increased mRNA for fibronectin and TGF-β, compared with the adjacent lung parenchyma. Procollagen-type...