as we have progressed technically, our tools for detecting subtle changes resulting from defective *CFTR* activity or expression have been refined, including the use of cell-specific knockouts, siRNAs, genomics, proteomics, and transcriptomic and genome editing. These tools have allowed the role of *CFTR*-deficient macrophages in the context of CF pathophysiology to be delineated (6, 15–17).

However, several questions remain: What do these changes really mean? How does a new “set-point” or altered XBP-1 activity affect the cell’s response to the environment? How important is the immune response beyond the contributions of the airway epithelium in CF? Is the contribution of the immunologic response to CF pathophysiology of a magnitude that it can be manipulated to significantly affect CF lung disease? Why is the concept of “specific” immune involvement so hard to embrace as an important focus? Whether you agree that intrinsic *CFTR* defects directly alter macrophage responses or that the observed differences are the result of the response to the altered *CFTR* milieu, what difference does it make? Therapeutic interventions aimed at XBP-1 or other molecules associated with sustaining the proinflammatory phenotype in the CF lung, in combination with antimicrobials and *CFTR* modulators, may provide improved resolution of infection and inflammation. The difference may not lie with only correcting *CFTR* function in airway epithelial cells and providing improved airway surface hydration, but also with providing supportive immunology, so that patients can aide in their own fight against infection and inflammation. This will ultimately improve quality of life, and hopefully survival, in patients with CF. ■

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Aerobic Glycolysis and the Warburg Effect An Unexplored Realm in the Search for Fibrosis Therapies?

Detailed discussion of the pathways involved in cellular metabolism is likely to evoke memories of undergraduate biochemistry. The enzymes and chemical intermediates required to generate

energy from carbohydrates and fatty acids were mapped out early in the 20th century by pioneers of metabolism, such as Hans Krebs. Until recently, these metabolic pathways have, as “known knowns,” been consigned to textbooks and have generated little, if any, research interest. This is, however, no longer the case. Driven by observations made in cancer cell biology, metabolism and the related field of metabolomics is now a rapidly growing area of

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interest, leading to therapeutic developments in both oncology and immuno-inflammatory diseases such as rheumatoid arthritis (1–3).

Glycolysis consists of a 10-step pathway through which glucose is converted to pyruvate with a net yield of two adenosine triphosphate molecules. In conditions of normoxia, almost all cells generate energy through the highly efficient mitochondrial-based process of oxidative phosphorylation (which generates 30–36 ATP molecules from a single glucose molecule) (4). Only during periods of hypoxia do cells usually revert to (anaerobic) glycolysis as their primary source of energy. However, as long ago as 1931, Otto Warburg was awarded the Nobel Prize for the observation that cancer cells, even in a normoxic environment, rely on glycolysis for the generation of energy, or the so-called Warburg effect (5).

The reason for this reliance on the relatively energy-inefficient process of aerobic glycolysis remains unclear, but the same mechanism has subsequently been observed in rapidly dividing normal cells and in lymphocytes and macrophages in immunologically driven disease (3). The large increase in glucose uptake necessitated by aerobic glycolysis underpins the method by which ^{18}F fluorodeoxyglucose-positron emission tomography enables detection and monitoring of many cancers. A growing body of evidence suggests that in cancer, aerobic glycolysis exerts effects that go far beyond energy production. Metabolites generated by glycolysis influence numerous aspects of cellular function, including proliferation, extracellular matrix production, autophagy, and apoptosis, as well as having effects on the behavior of bystander cells (1–3, 5).

So how might any of this be relevant to idiopathic pulmonary fibrosis (IPF)? On the basis of histological and radiological observations, areas of honeycomb change in IPF are generally considered to be “burnt-out” regions of established scar tissue. The relatively sparse fibroblastic foci seen in usual interstitial pneumonia are, in contrast, thought to represent the regions of “active” disease and to be the principal source of freshly produced collagen and extracellular matrix (6). However, established honeycomb-fibrosis demonstrates marked activity on ^{18}F fluorodeoxyglucose-positron emission tomography, suggesting that contrary to the prevailing dogma, fibrotic lung tissue is highly metabolically active (7, 8). In keeping with this observation, there is also evidence pointing to up-regulation of glycolytic pathways in IPF. Kottmann and colleagues have shown that lactic acid is increased in the lungs of individuals with IPF when compared with disease-free controls (9). Furthermore, this excess of lactic acid (produced through the conversion, by lactate dehydrogenase, of pyruvate) plays a role in activating the profibrotic cytokine transforming growth factor (TGF)- β .

In this issue of the *Journal*, Xie and colleagues (pp. 1462–1474) have now taken these observations a step further by demonstrating that human lung fibroblasts (differentiated into myofibroblasts through exposure to TGF- β) exhibit high levels of aerobic glycolysis that act to facilitate progression to, and maintenance of, the transformed myofibroblast state and that enhances contractility and cellular migration (10). This up-regulation of glycolysis and the consequent downstream effects are even more marked in IPF-derived lung myofibroblasts. Importantly, Xie and colleagues demonstrate that the observed effects of glycolysis on fibroblasts do not appear to be inherently dependant on the relatively small amount of energy generated by this pathway. Instead, aerobic glycolysis drives the production of succinate, which acts, independent of hypoxia, to stabilize the transcription factor, hypoxia-inducible factor 1, which in turn promotes TGF-

β -induced fibroblast-to-myofibroblast transformation. Importantly, partial blockade of glycolysis with the 6-phosphofructo-2-kinase/fructose-2,6-bisphosphatase (PFKFB)-3 inhibitor 3-(3-pyridinyl)-1-(4-pyridinyl)-2-propen-1-one reduces the profibrotic effects of TGF- β *in vitro* and attenuates the development of fibrosis in both the bleomycin- and TGF- β -induced murine models of fibrosis.

Although these data highlight a hitherto unrecognized pathogenic mechanism in IPF, several questions remain unanswered (10). First, the mechanisms underpinning activation of aerobic glycolysis in fibroblasts, together with the further up-regulation of these pathways seen in IPF fibroblasts, remains to be explained. Second, Xie and colleagues focused the majority of their experiments on a single enzyme (PFKFB3) and a single metabolite (succinate) in the glycolytic pathway. The role played by the myriad other enzymes and metabolites involved in glycolysis remains to be explored. Third, the authors have assumed that the observed increase in succinate arises because of activation of the tricarboxylic acid cycle (part of the mitochondrial oxidative phosphorylation pathway). However, in other settings, enhanced aerobic glycolysis results in down-regulation of oxidative phosphorylation resulting from inhibition of this pathway by lactate (11). Succinate synthesis is therefore likely to be driven by other mechanisms, including via anaplerosis from glutamine conversion to α -ketoglutarate and through a γ -aminobutyric acid shunt (3, 12). Better understanding of the mechanisms driving succinate accumulation in fibroblasts may identify further potential therapeutic targets. Finally, the authors have focused on the intracellular consequences to fibroblasts of enhanced glycolysis. In the context of cancer, it has been shown that aerobic glycolysis by stromal fibroblasts results in secretion of the energy-rich metabolites lactate and pyruvate (13). These are then taken up by tumor cells, leading to an up-regulation of oxidative phosphorylation. This phenomenon, termed the reverse Warburg effect, drives epithelial cancer proliferation and generates tissue-damaging oxygen free-radicals (13). The reverse Warburg effect could also be an important pathogenic mechanism in IPF, with secreted metabolites potentially influencing the behavior of various cell types, including epithelial cells and macrophages, and even possibly the resident microbiome (14, 15).

These issues notwithstanding, Xie and colleagues have identified a novel disease mechanism contributing to the development of fibrosis, and in doing so, they have highlighted a potential new approach for treating fibrotic disease (10). It seems likely that ongoing advances in metabolomics and the potential to integrate any findings with genetic, transcriptomic, and proteomic data will lead to a rapid expansion in understanding the role played by metabolic abnormalities in the pathogenesis of disease. It is possible, therefore, that knowledge gained through pioneering biochemical experiments performed during the early 20th century will, almost 100 years on, facilitate the discovery of innovative treatments for fibrosis. ■

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Personalizing Therapy in Idiopathic Pulmonary Fibrosis: A Glimpse of the Future?

Over the past few years, advances in genetics and therapeutics have transformed our understanding of idiopathic pulmonary fibrosis (IPF) and brought new hope for this debilitating disease. In 2014, based on the results of concurrently published phase III clinical trials, the first IPF treatments were approved by the U.S. Food and Drug Administration. These two medications, pirfenidone (1) and nintedanib (2), target fibroblasts and have been shown to slow disease progression. In parallel, the PANTHER-IPF (Prednisone, Azathioprine, and N-acetylcysteine: A Study That Evaluates Response in Idiopathic Pulmonary Fibrosis) trial (3) provided convincing evidence that traditional immunosuppressive “triple” therapy for IPF with prednisone, azathioprine, and N-acetylcysteine (NAC) was not effective and likely harmful for patients with IPF. In contrast to

findings from the earlier IFIGENIA (Idiopathic Pulmonary Fibrosis International Group Exploring N-acetylcysteine I Annual) study (4), the NAC versus placebo arms of the PANTHER trial (5) found no benefit to NAC treatment.

During this time period, genome-wide linkage and association studies, in addition to next-generation sequencing technologies, have identified rare and common genetic variants that account for at least 30% risk of IPF (6) and may predict disease outcomes. In particular, several common polymorphisms on chromosome 11p15 were identified as risk factors for IPF, including a single-nucleotide polymorphism (SNP) in the promoter for the gene encoding Mucin 5B (*MUC5B*) (7) and SNPs in the Toll-interacting protein (*TOLLIP*) locus (8). Both the *MUC5B* (9) and *TOLLIP* (8) risk alleles were associated with disease outcomes, although, surprisingly, patients with IPF with these risk alleles had better outcomes than subjects without the risk alleles.

With increasing understanding of the genetic heterogeneity of IPF, natural next questions are: (1) Might genetic factors predict response to IPF therapy? and (2) Could a subset of patients with

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