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Commentary

Summary of Drug Needs in Cystic Fibrosis[1]

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At the pathophysiologic level, cystic fibrosis (CF) appears to reflect epithelial dysfunction in target organs. Basic science studies have identified defects in the regulation of transepithelial ion movements across affected epithelia in this disease that may account for many of the clinical manifestations of this disorder. Major efforts are being devoted to understanding the CF defect more completely at the biochemical level and, ultimately, the molecular level. It is possible that a number of reagents currently may be available that can assist in these efforts. Potentially useful reagents have been broken down into two categories: (I) compounds/vectors that may be useful for basic studies of the biochemical and molecular effects of cystic fibrosis to provide, in the short term, important insights into the pathophysiology of CF and, in the longer term, therapeutic windows for treating this disease; and (II) compounds/devices that can immediately increase the efficacy of treatment of cystic fibrosis.

I. Compounds/vectors for use primarily in *in vitro* systems to investigate the basis of cystic fibrosis epithelial defects at the cellular and biochemical level
   A. Compounds that affect cytoplasmic membrane ion conduction. Cystic fibrosis epithelia exhibit abnormalities in channel activation: Cl^- channels remain inactive, while Na^+ channels display excessive activation. The following classes of compounds would be useful for modeling purposes in normal epithelia and for exploring modes of bypassing channel activation defects.
      1. Specific blockers of ion channels that are affected in cystic fibrosis, e.g., sodium and chloride channels. Whereas amiloride may serve as a relatively specific and useful blocker of the sodium channel, blockers of chloride channels are few. These blockers would be extremely useful in studies designed to model CF defects. Cl^- channel blockers may be sought by companies interested in inhibiting symptoms of secretory diarrheas.
      2. Monovalent ionophores. A potentially very useful compound would be a relatively selective chloride ion ionophore. This agonist might allow translocation of chloride across CF membranes that are otherwise chloride impermeable and, consequently, test the notion that rectification of this defect may be therapeutic.
   B. Agents that modulate intracellular calcium. It appears that in some CF target epithelia, cAMP and protein kinase C-dependent mechanisms for activating ion channels are defective, whereas parallel Ca^{2+}-dependent mechanisms remain effective. Consequently, a potential mode of therapy is to raise cytosolic calcium in affected cells for extended periods. Several classes of compounds would be worth exploring in this endeavor.
      1. Calcium-ATPase inhibitors
      2. Sodium-calcium exchange inhibitors
      3. Specific calcium ionophores
   C. Specific inhibitors of intracellular regulatory enzymes. Obviously, when one deals with defects in intracellular regulation, complex processes with many interactions are involved. With regard to CF, a number of specific inhibitors would be quite useful for exploring the function in normal cells of pathways potentially defective in CF and for attempting to rectify CF defects by redressing regulatory imbalances in CF cells. Most of the emphasis on the regulatory defects in cystic fibrosis has focused on phosphorylation systems that may be affected by this disease. Consequently, the agents that would be of most use fall into the following classes.
      1. Inhibitors of calcium/calmodulin-dependent kinases
      2. Inhibitors of protein kinase C
      3. Inhibitors of protein kinase A (cAMP-dependent protein kinase)
      4. Phosphatase inhibitors
   D. Compounds that affect intracellular sulfate (SO_4) metabolism. Recently it has been discovered that the anion transport defect in CF epithelia affects not only monovalent anions (chloride) but also divalent anions, e.g., sulfate. An interesting consequence of the abnormal sulfate metabolism in cystic fibrosis cells is that the degree of sulfation of the carbohydrate components of the cell surface glycocalyx may be abnormal. Abnormalities of the glycocalyx may potentially change the binding sites for microorganisms on the airway cell surface. Compounds that might affect the

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degree of sulfation of this important component of cell surface defense include the following.

1. Inhibitors of cell sulfate uptake translocation pathways
2. Specific inhibitors of sulfotransferases

E. Compounds that affect oxidoreductase activities (redox potential) of epithelial cells. A number of studies have recently indicated that CF epithelial cells exhibit abnormal rates of oxidative metabolism. Consequences of this increased metabolism on the electron transfer mechanisms are not known. Useful agents for such studies would involve the following.

1. Compounds that monitor oxidative activity intracellularly.
2. Products that alter cellular redox potentials.

F. Vehicles that can deliver hydrophilic compounds to the intracellular compartment. It is highly probable that an initial outcome of the identification of the cystic fibrosis gene product will be an attempt to introduce the normal gene product into the CF cell. It is likely that this product will be a large, hydrophilic protein, and consequently, the introduction of such a material to the cell compartment will present a formidable challenge. Therefore, strategies and/or devices designed to deliver proteins to the cell interior, presumably as delivered by aerosol directly to affected cells in the lung, should be contemplated. Such devices include the following.

1. Liposomes, antibody and/or charge specific substances.
2. Molecules for which there are naturally occurring receptors on cell surfaces that may serve to trigger endocytosis of therapeutic compound–protein complexes into the cell.

G. Vectors for transforming epithelial cells. The supply of epithelial cells for study from both normal patients and cystic fibrosis patients is severely limited. Consequently, the ability to extend the proliferative state of cultured epithelial cells by the introduction of activated proto-oncogenes potentially offers a useful mechanism for increasing the mass of cell material available for study. Particularly useful are the following.

1. Oncogenes, preferably under-temperature sensitive or metallothionine promoters, that would potentially be effective in transforming human epithelial cells.
2. Vectors that selectively target airway epithelial cells for gene introduction. Such receptors might include the naturally occurring viral receptors that may be expressed on airway cells, e.g., for influenza, adenovirus, and respiratory synticial virus.

II. Agents with promise to improve the clinical care of CF patients

A. Improved antibacterial agents. Despite the great advances in prolonging the life span of cystic fibrosis patients with aggressive antibiotic therapy, the problem of persistent and often resistant organisms, typically of *Pseudomonas* and *Staphylococcus* species, is the most pressing clinical problem of cystic fibrosis. Consequently, efforts should immediately be focused on improved killing of these organisms, thus decreasing the chronic bacterial burden in the lungs of these patients. Strategies to be explored include the following.

1. Improved antibiotics that would focus specifically on *Pseudomonas aeruginosa* and *Pseudomonas cepacia*. Effective antibiotics that can be given by the entire spectrum of routes, i.e., intravenous, aerosol, and oral.
2. The possibility of vaccination approaches to inhibit the severity of the staphylococcal and/or pseudomonal infections in the lungs.
3. Novel therapeutic strategies that might focus on prevention of bacterial binding and/or attachment to the epithelium of cystic fibrosis airways.

B. Agents that prevent lung transplant rejection. A form of therapy for end-stage lung disease in cystic fibrosis is the heart–lung transplant. It appears clear from the initial studies of 60 transplant patients worldwide that the problem of chronic pulmonary rejection has not been solved. Two routes for improving the outcome of these patients with regard to rejection are the following.

1. Improved systemic immunosuppressive agents
2. Development of immunosuppressive agents that could be delivered directly to the lung via the aerosol route.

C. Improved anti-inflammatory agents. Preliminary clinical studies indicated that anti-inflammatory agents may reduce the lung deterioration seen in patients chronically colonized with pathogens. Several large-scale clinical trials are now under way.

D. Improved aerosol delivery devices. It is probable that a number of potentially useful therapeutic agents can be delivered to the lungs of cystic fibrosis patients via the aerosol route. A spectrum of agents that can be delivered by this route includes antibiotics, steroids, diuretics, and, conceivably in the future, vehicles designed to dose the intracellular compartment and/or effect genetic therapy. The techniques for delivering aerosols to the lung require considerable refinement. Important improvements in existing devices and development of novel devices include the following.

1. Aerosol generation systems. A major goal is to improve the delivery system for dry powder aerosols.
2. Scrubbers/impacters. Many of the current aerosol delivery devices generate large particles that impact on the oropharynx and are swallowed. For many of the compounds of interest, absorption of drugs may be undesirable because of the capacity to induce side effects via the oral route and/or to limit the ceiling concentrations of drugs delivered to the lung. Consequently, devices designed to remove

the particles that impact on the oropharynx are desirable.

3. Devices to sustain delivery of drugs on airway surfaces. For convenience to patients, it is desirable for drugs delivered via the aerosol route to be taken no more than twice daily. Therefore, a device or devices that retain the drug in the airway compartment and/or release it slowly into this compartment are desirable. Potential candidates are liposomes and/or cross-linked albumin microspheres.
4. Devices to impart energy to inhaled aerosols. Patients with lung disease have low inspiratory flow rates and, consequently, impart relatively little energy to particles that are being inhaled into the lung. The result is central deposition of drug and underdosing of the peripheral lung compartment. A number of ways to impart more energy to particles and increase the homogeneity of deposition to the lung, e.g., forced random noise techniques, should be explored.