Drug: a medicine or other substance which has a physiological effect when ingested or otherwise introduced into the body

Defining the term drug turns out to be surprisingly difficult. A quick Google search afforded me with the definition above. It doesn't take long however to find fault with this definition, which in my mind would define chocolate ice cream as a drug. The Food and Drug Administration includes as part of their definition a somewhat similar concept, qualified to ease my ice cream concern.
articles (other than food) intended to affect the structure or any function of the body of man or other animals.

I think most people however, would be most comfortable with the following component of the FDA's definition of drug:
*articles intended for use in the diagnosis, **cure, mitigation, treatment** or prevention of disease in man or other animals.*

The field of science that specializes in the all aspects of drugs including their uses, limitations, modes of action, and how to discover more and better drugs is referred to as pharmacology; and industry that specializes in manufacturing, packaging, and marketing drugs, the pharmaceutical industry. A recent YouTube video highlighting an increased interest of the pharmaceutical industry in identifying drugs for rare diseases like DBA seemed like a natural call for a newsletter article on the relationships between the pharmaceutical industry, academic researchers, and private foundations and advocacy groups.

Pharm 101

Drug Targets
Drugs have their effects via their interactions with molecules within the body. These molecules are referred to as drug targets. Sometimes these targets are products of the genes affected in the disease. A great example of this concept is the drug Kalydeco developed to treat cystic fibrosis. Kalydeco targets the cystic fibrosis transmembrane regulator (CFTR), a chloride transporter found on the surface of certain epithelial cells in the body, particularly those in the lung. Kalydeco was developed through a partnership between the Cystic Fibrosis Foundation and the pharmaceutical company Vertex Pharmaceuticals. As part of this partnership, the Cystic Fibrosis Foundation invested $75 million into research and development at Vertex with the specific goal of identifying a drug to treat cystic fibrosis.

This investment was a huge risk for the CF Foundation because to put all their eggs in the Vertex basket (so to speak) they had to cut back on other lines of investigation. The risk was mitigated somewhat by the payoff: big risk, big payoff. With Kalydeco's recent approval by the FDA for treating certain forms of cystic fibrosis, this partnership can clearly be viewed as a resounding success story. In a monetary sense, this success story could be kicked up a notch to absolutely phenomenal, as the Cystic Fibrosis Foundation has recently sold its royalty rights to Kalydeco for 3.3 billion dollars.

The limits of the Kalydeco success story for patients reside in the fact that this drug targets only a small subset of genetic defects found in the CTFR gene and does not work for, F508del, the mutation found in the majority of cystic fibrosis patients. However, with 3.3 billion dollars to invest in additional research, the future certainly looks bright for the development of further drugs to treat a wider spectrum of CF patients.

Potentially lost in the Kalydeco success story is that the success of this drug was built on the shoulders of a tremendous amount of basic research on cystic fibrosis funded by private foundations and various government agencies, including the discovery of the CTFR gene in 1989. Without knowledge of the structure and function of the CTFR protein and countless other studies on fundamental approaches to gene discovery, it is doubtful that Kalydeco's success could have been realized.

Similar groundwork work has been, and continues to be, laid down for the DBA field. Both private foundations and government agencies have supported research on DBA and tremendous progress has been made in the past 10 years on the genes affected in DBA and the pathogenic processes

involved. These studies have led to the identification of numerous potential targets that are being exploited in the development of drugs to improve the lives of DBA patients and their families.

The Drug Screen

Early stages of drug development typically center on what is called a drug screen. 160,000 compounds were analyzed as part of a high throughput screen used to discover the lead drug, which was further modified to produce Kalydeco [1]. A critical feature in drug discovery is the nature of the high throughput screen. When testing 160,000 different compounds, the nature of the screen is critical. A screen that involves multiple convoluted steps, difficult measurements, or expensive reagents (including animals) makes screening hundreds of thousands of compounds unfeasible for many diseases. With significant funds in hand however, even difficult screens can be pulled off by shear brute force.

The screen used in Kalydeco development was one of these brute force screens. The screen involved placing a defective CTFR gene into mouse cells that do not normally express CTFR and then analyzing different compounds for their ability to rescue CTFR function using a change in cell fluorescence as a means to monitor CTFR function. Having identified lead compounds in this relatively crude system, investigators at Vertex then began testing the compounds in the more natural setting of cells cultured from the airways of CF patients.

Small Molecules (code for a drug with useful pharmacological properties)

For a drug to be effective it has to reach its target in the human body. To get into the human body a drug could be ingested orally or avoid the harsh environs of the digestive tract via injection. Most of us I suspect would prefer the oral route of drug delivery. For this to be effective however, the drug must avoid being destroyed in the strong acidic environment of the stomach or being degraded by the various enzymes released in the intestine, and if the drug is fortunate enough to get this far intact, it needs to be recognized and taken up by the cells lining the GI tract for entry into the circulation and delivery to other parts of the body. Drugs also must be relatively inert chemically, at least in these early stages after ingestion to avoid damaging cells within the GI tract and chemically inactivating the drug. Finally, once within the body, the drug has to avoid being recognized as foreign and either taken up by the liver for destruction, or being recognized by the immune system, or even being peed out via the kidney. Many drugs identified initially in simpler cell or molecule-based screens fail to pass muster when it comes to testing in more complex organismal settings.

In general, relatively small molecules tend to be more likely to have these types of useful pharmacological properties, than let's say a drug based on a larger compound like a protein. An example of a protein based drug would be Sotatercept, discussed in a recent newsletter article. It is not that such a drug may not be useful because of its size, it is just that when using such a drug (another example being insulin) the drug has to be injected into the body, rather than be delivered by oral ingestion.

The initial compound identified in the Vertex screen for compounds that rescued CFTR defects had to undergo numerous rounds of chemical refinement to increase its potency and improve it pharmacological properties [2]. This type of refinement process is where the pharmacological industry tends to excel because of the diverse sets of skills needed for the chemical refinement and in drug disposition in organismal settings.

Also included in these later stages of drug development is toxicity testing. As mentioned above, drugs elicit their effects by interacting with a target. Sometimes, however, drugs also interact with other molecules in the body classified as off target effects, more commonly known as side effects. Here again, drugs can fail after millions of dollars worth of investment in earlier stages of development, which is one of the reasons pharmaceutical companies provide for the high costs of drugs that eventually make it to market.

While perhaps not on the scale of screen undertaken by Vertex, the DBA Foundation is supporting multiple laboratories screening for drugs that may be effective in DBA patients (for details, see previous newsletter articles or our website: http://dbafoundation.org/category/research/). Dr. Harvey Lodish at MIT has taken the approach of screening for drugs that synergize with steroids in enhancing

erythropoiesis in cells derived from the mouse fetal liver. The idea behind this screen is that compounds that synergize with steroids in these normal mouse cells may serve as lead compounds for the development of drugs that also synergize with steroids in the context of a DBA patient. The combination of such a drug with steroids could allow physicians to use reduced levels of steroids or it may be possible to have such a drug completely replace steroids. The hope here is to obtain the same clinical benefit that patients currently receive with steroids, with fewer steroid-related side effects. In an alternative screen, Johan Flygare is looking for compounds that rescue the hematopoietic phenotype of the Swedish mouse model of DBA. Additionally, Dr. David Bodine is funded to screen for small molecule therapeutics that increase ribosomal protein expression in human cell lines, a strategy that could also lead to more effective DBA treatments. Finally, with the recent creation and characterization of induced pluripotent stem cells from DBA patients by groups at the Children's Hospital of Philadelphia, a resource has been created that should provide fertile ground for future drug screens in a more refined setting using human cells that are more closely related to the targets within the body [3].

Repurposing Drugs

As noted above, one of the huge challenges in drug development are the difficulties encountered when taking the drug from the lab bench to organismal studies and human trials. The costs incurred in the refinement of lead compounds and the ultimate losses incurred when a drug fails in late stages of development make drug discovery a long and costly process. For this reason, there is much interest in the concept of repurposing currently used drugs for other purposes. The idea here is that when setting up a more limited screen on a shoestring budget, it might first be best to screen drugs currently used for other purposes for efficacy in a rare disease like DBA. Such drugs have already been formulated to maximize their pharmacological properties, further, their side effects in human populations are known and can be taken into consideration early in the drug development process to avoid costly investments or serious adverse effects. Such an approach has been taken in a separate screen undertaken in the Lodish laboratory, also supported by the DBA Foundation.

I hope you get a sense from this newsletter article that perhaps not on the scale of that discussed for Cystic Fibrosis Foundation and Vertex Pharmaceuticals, progress is being made in several screens for drugs to improve the lives of DBA patients and their families through support provided by your generous donations and fund raising efforts.

1. Van Goor, F., et al., *Rescue of DeltaF508-CFTR trafficking and gating in human cystic fibrosis airway primary cultures by small molecules.* Am J Physiol Lung Cell Mol Physiol, 2006. **290**(6): p. L1117-30.
2. Hadida, S., et al., *Discovery of N-(2,4-di-tert-butyl-5-hydroxyphenyl)-4-oxo-1,4-dihydroquinoline-3-carboxamide (VX-770, ivacaftor), a potent and orally bioavailable CFTR potentiator.* J Med Chem, 2014. **57**(23): p. 9776-95.
3. Garcon, L., et al., *Ribosomal and hematopoietic defects in induced pluripotent stem cells derived from Diamond Blackfan anemia patients.* Blood, 2013. **122**(6): p. 912-21.