



Enter Sofosbuvir: The Path to Curing HCV

Michael J. Sofia

The decision to leave the security of a large pharmaceutical company and jump into the unknown of a small biotech called Pharmasset with 15 staff members, limited capabilities, no labs, and little money would to many seem a foolish decision. In fact, many advised me to not make the move. But the desire and excitement to blaze a new trail where none had been, to build a research team where one barely existed, and to work in an exciting area that was just beginning to develop outweighed the thoughts of risk and uncertainty. This is a story of how such perhaps lofty aspirations turned into the most satisfying chapter of my career and how a collaborative endeavor resulted in a drug that went beyond our hopes and expectations.

In 2005, Pharmasset had three research and development programs for antiviral drugs, one of which targeted the hepatitis C virus (HCV). Hepatitis C is an insidious disease that infects the liver and progresses over time to chronic liver disease, liver fibrosis, liver cirrhosis, and eventually to liver cancer. Approximately 170 million individuals worldwide are infected with HCV, and 700,000 die of HCV-related liver disease each year. The standard of care for treating HCV infection was to harness the responses of the immune system while targeting aspects of viral reproduction with a combination of interferon and a nucleoside inhibitor ribavirin for 48 weeks. This lengthy treatment regimen produced severe side effects, primarily due to the effects of interferon, such that many patients did not complete therapy and it only delivered a modest cure rate in a subset of HCV genetic variants. Consequently, there was a significant effort to find new therapies for HCV that would ultimately eliminate the need for interferon. At the time, there were a large number of biotech and pharma companies developing drugs against HCV. Several of the more advanced programs in clinical development were investigating HCV protease inhibitors, which showed promise but only when combined with interferon and ribavirin. They also had serious drawbacks: they were associated with many undesired side effects, were limited in their ability to reduce viremia within the 48-week period of treatment, and lacked the ability to

“Approximately 170 million individuals worldwide are infected with HCV, and 700,000 die of HCV-related liver disease each year.”

Discovery research leadership team at Pharmasset (L to R: Michael Sofia, Michael Otto, and Phil Furman).

cover a broad diversity of HCV genotypes. Also of major concern was the rapid emergence of drug resistance.

When I arrived at Pharmasset, their nascent HCV program was focused around a nucleoside analog, PSI-6130. Nucleoside analogs are versions of the natural building blocks that make up the DNA and RNA genetic code of living organisms. This cytidine nucleoside analog, which has a unique 2'- α -fluoro-2'- β -C-methyl chemical modification, targeting the HCV RNA-dependent RNA polymerase to inhibit synthesis of viral RNA, demonstrated promising activity in an assay that tested the ability of HCV to replicate in cells grown in culture, seemed to be well tolerated in cells and animal studies, appeared to have a broad genotype coverage, and exhibited a high barrier to resistance. The challenge with this molecule was that it exhibited modest potency; only a small proportion of it was available in the body when taken by mouth (a drug parameter called bioavailability) and generated a significant amount of an inactive uridine metabolite, thus reducing the amount of available active drug. An attempt to solve this problem led to the development of prodrugs of PSI-6130. Prodrugs can be thought of as a "Trojan Horse" form of the parent drug. By masking the parts of the drug that are believed to be responsible for its inability to get into the body, it is possible to change the way the drug looks to cells, and it can therefore be absorbed more efficiently. The early clinical data on the prodrug named RG7128 was indeed quite encouraging. RG7128 demonstrated efficacy against HCV either when administered alone or in combination with interferon + ribavirin. This coupled with the lack of observed resistance and the absence of adverse events associated with RG7128 increased our confidence in this approach. We were particularly encouraged by the positive results achieved by RG7128 in combination with an HCV protease inhibitor but without interferon—the first hint in human clinical trials that an interferon-free direct-acting antiviral drug combination had promise. However, this nucleoside prodrug also had drawbacks: it still had only modest potency, grams of this agent needed to be administered to patients several times a day to see a clinical effect, and the undesired uridine metabolite was still produced in significant quantities. Thus, although RG7128 provided a proof of concept for the use of 2'- α -fluoro-2'- β -C-methyl nucleoside and continued to progress in clinical trials, several of us at Pharmasset believed that we needed to find an even better drug if we were going to achieve the ultimate goal of developing a cure for HCV. Both outside and even within the company, this notion was not universally accepted. Some believed we had the drug in RG7128 and were not contemplating something better. This limited enthusiasm for furthering nucleoside drugs also came from the perception that other compounds, such as HCV protease inhibitors, were further along in clinical studies. And there was always the question of whether a nucleoside would be potent enough to compete with these other agents. At the time, the clinical data did not support this hope.

I strongly felt that, to make a better nucleoside drug, we needed to tackle, among other factors, the issue of its potency while maintaining its good characteristics, such as its pan-genotypic character, high barrier to resistance, and high selectivity. Once-a-day dosing was also strongly desired, and selective targeting of the liver where HCV resided was seen as critical but aspirational. The question became how to identify such a drug.

My team had already carried out extensive structural modifications around the 2'- α -F-2'- β -C-methyl nucleosides and was not able to identify any analog that was better than PSI-6130, so the next step was not obvious. However, understanding its metabolism, based on careful studies done by my colleague Phil Furman and his group, turned out to be critical in determining how we might solve the potency and drug duration issues. Indeed, he and his group became an important part of our success.

As with virtually all nucleoside analog drugs, the nucleoside itself is not the form that inhibits the viral polymerase. The nucleoside must be converted in the target cell into the nucleoside 5'-triphosphate, and it is this metabolite that inhibits the viral polymerase by ultimately behaving as a substrate and causing a chain elongation termination event. One possibility for its low activity was that the PSI-6130 triphosphate active metabolite had a short half-life in primary human hepatocytes and didn't provide a sustained high concentration. The metabolic studies had shown us that, not only does PSI-6130 get converted to its active 5'-triphosphate, but in fact, a 5'-monophosphate metabolic intermediate was further metabolized into a uridine

"Although the logic seemed sound, at least to me, there was considerable skepticism amongst most of those whose opinion was sought about the idea as to whether or not this would work."

5'-monophosphate derivative, which in turn produced an active 5'-triphosphate, which had a long half-life in primary human hepatocytes, potentially allowing for increased potency and once-daily dosing. Thus, understanding the metabolism of PSI-6130 indicated that there is a metabolite—the uridine triphosphate—with the exact properties that we were seeking for an inhibitor of HCV polymerase. Unfortunately, however, assays in cells showed that just adding a uridine nucleoside analog was not sufficient to trigger the formation of the uridine triphosphate, as there was a block in the metabolic conversion of the uridine nucleoside analog to its 5'-monophosphate derivative and therefore the target triphosphate was never produced. To overcome this pathway block, we would need to deliver into the cell and ultimately into a human a nucleoside 5'-monophosphate so that it could then proceed onto the triphosphate form. The dilemma was that nucleoside 5'-monophosphates are highly charged and would never get into cells, let alone pass through the gastrointestinal tract. Moreover, monophosphates are chemically and enzymatically unstable. The path forward required us to develop a way to mask the nature of the phosphate group in such a way that it could survive the gastrointestinal tract, get absorbed, and get to the liver intact, followed by release of the monophosphate in hepatocytes. The concern was that this had never before been achieved in an in vivo setting, let alone in a human. In addition, we had the high ambition of attempting to selectively target the liver, thus producing high liver exposure relative to other parts of the body to minimize any off-target adverse effects—again, a task never attempted with a prodrug of a nucleoside monophosphate or, in fact, any other drug type.

One of the “a-ha” moments came when it dawned on me that, if we could leverage liver first-pass metabolism (the liver, which is the most metabolizing organ in the body, is the first organ that anything—food or drug—sees when it gets absorbed into the body from the gut and before it circulates throughout the rest of the body) to selectively de-mask the phosphate group, we might be able to achieve selective presentation of the drug in the liver. With that concept in mind, we chose the phosphoramidate promoietty construct, in which the phosphate group is modified and unrecognizable as such until it is metabolized in the liver, to see if we could build a molecule that achieved our objectives. Although the logic seemed sound, at least to me, there was considerable skepticism among most of those whose opinion was sought about the idea as to whether or not this would work. It was simply believed that no one had ever been able to achieve the delivery of a nucleoside phosphate in humans before, so why should it work now? With a strong personal belief in the idea, the first phosphoramidate prodrug of the uridine nucleoside was designed and synthesized, and when tested in the whole-cell HCV replicon assay, antiviral activity was observed. However, this was far from being a drug. Because there was no precedent for what we were trying to achieve, we realized that we needed to establish an in vitro and in vivo screening paradigm that would allow us to select the best drug candidate from a large number of prodrug analogs. Setting up systems for screening a drug working on new principles took time, but after a series of comprehensive screens, we ultimately chose a molecule called PSI-7851 as a clinical candidate.

PSI-7851 possessed all of the characteristics for which we were hoping, and consequently, it was taken into the clinic to demonstrate proof of concept in humans. Although we were very excited about the potential for PSI-7851, my excitement was somewhat tempered from many years doing drug discovery. In drug discovery, you can do all the preclinical work possible to validate the potential of a drug candidate, but the ultimate validation is efficacy and safety in humans, and too many times the preclinical work does not translate into the clinic.

In a phase 1 clinical study in HCV genotype-1-infected patients, PSI-7851 provided the proof of concept we had hoped for and validated the concept that this drug could get to the liver and deliver the monophosphate that ultimately progressed onto the triphosphate and inhibited HCV. This was the first time that a phosphate prodrug was shown to have efficacy in a human clinical setting. This was a big achievement in its own right, but it was not the end to the story because PSI-7851 was, in fact, not the drug we wanted. The problem with PSI-7851 came down to chemistry. The chemistry we used to build PSI-7851 had an inherent flaw. It produced a mixture of isomers, compounds with the same formula but a different arrangement of atoms, which conveyed different properties. Clearly, an ideal drug would need to be a single isomer to ensure that our drug acted in a reliable and specific way. But at the time, no chemical methods existed to easily separate the two isomers, let alone selectively synthesize the single isomer. Undaunted, we

“Sofosbuvir has been described as a miracle drug, and its contribution to the cure of HCV is regarded as one of the most significant public health accomplishments of our generation.”



The chemistry team at Pharmasset: January 2012.

were ultimately able to not only develop a method to crystallize the more potent single isomer, PSI-7977, and obtain its X-ray structure, which gave us insights into the way the structure of the molecule informed its action, but we also developed a novel previously unknown method to chemically synthesize PSI-7977. PSI-7977 was ultimately named sofosbuvir. Sofosbuvir was taken into phase 2 clinical trials and showed exceptional potency in combination with interferon/ribavirin. However, it wasn't until a landmark clinical study called "Electron" that the full potential of sofosbuvir was realized. In this study of HCV genotype 2 and 3 patients, the interferon-free combination of sofosbuvir and ribavirin dosed for 12 weeks resulted in a 100% cure rate. This result was a game-changer for HCV patients because for the first time an interferon-free regimen demonstrated high cure rates, was well tolerated, and exhibited no drug resistance. Sofosbuvir + ribavirin interferon-free HCV cure therapy was first approved by the US FDA on December 6, 2013. Sofosbuvir has gone on to become the backbone of combination regimens that include ribavirin, HCV protease inhibitors, and HCV NS5A inhibitors for treatment of HCV infection of all genotypes and for a broad patient population. To date, the real-world experience with sofosbuvir-based regimens has mirrored the clinical trial experience, and it is estimated that more than 800,000 patients have been cured of their HCV and have gone on to live normal lives without the specter of debilitating chronic liver disease, liver cirrhosis, or liver cancer.

During the entire discovery and development effort that led to sofosbuvir, I was completely immersed in following the science and solving the many problems that were faced along the way. It wasn't until I attended the US FDA advisory board meeting that reviewed sofosbuvir and made the unanimous recommendation that sofosbuvir should be approved as a curative therapy for HCV patients that I truly realized the impact this drug was going to have for patients suffering from



Dr. Onaiwu Ogbomo (L), cured by sofosbuvir-based regimen, and Michael Sofia (R) at the FDA Advisory Board Meeting on October 25, 2013.

HCV. It was during the public comment session at this meeting that the story of Dr. Onaiwu Ogbomo would sum up the true impact of sofosbuvir. He was a young boy when he contracted HCV, probably from a vaccination program in his home country, Nigeria. He came to the United States to teach as a college professor after earning his Ph.D. in Canada. His liver disease got progressively worse until his doctor recommended he get a liver transplant. He declined a transplant because he feared that he might die before he saw his children graduate from college. His children graduated, and he subsequently survived a liver transplant only to be given the bad news that his HCV had reoccurred, which frequently happens to transplant patients. His doctor had no more options for him and advised him to get his affairs in order. Soon after, he was contacted by his doctor, informing him that there was a clinical trial for a drug sofosbuvir and asking if he was interested in participating. He joined the clinical trial, and 36 weeks later he was cured of his HCV. On October 25, 2013, he stood before the FDA advisory panel to share his moving story. Today, there are thousands of stories like Dr. Ogbomo's, highlighting how sofosbuvir has given HCV patients a new lease on life. Sofosbuvir has been described as a miracle drug, and its contribution to the cure of HCV is regarded as one of the most significant public health accomplishments of our generation. I am humbled and grateful to have played such a role in its discovery and development.

Looking back, it seems that my story came full circle, as I have in the meantime moved on to pursuing new challenges in viral hepatitis drug development, away from Pharmasset: the pursuit of a cure for chronic hepatitis B virus infection. I carry with me the knowledge that the extraordinary success of sofosbuvir would not have happened if it weren't for a talented group of individuals who embraced an idea and helped make it a reality. HCV patients and I will forever be indebted to them for their dedication and hard work.