This paper is available online at www.jbc.org
THE JOURNAL OF BIOLOGICAL CHEMISTRY VOL. 284, NO. 52, pp. 35997–36006, December 25, 2009

From Molecular Biology to Geology: A Surprising Trajectory

Published, JBC Papers in Press, October 23, 2009, DOI 10.1074/jbc.X109.076836

John Abelson

From the Department of Biochemistry and Biophysics, University of California, San Francisco, California 94131

It has been nearly 50 years since I started doing molecular biology as a graduate student in the Department of Biophysics at The Johns Hopkins University. I am still doing molecular biology, but I now find myself just as interested in geology; and although I will never really be a geologist, I have been on some fascinating field trips in the past eight years to South Africa, Namibia, Oman, Australia, the Bahamas, and the American West. I am even coauthor of a paper in geology (1), so I will reflect in this article how this came to pass.

In 1965, when I left Johns Hopkins to begin my postdoctoral position in Cambridge, England, there were almost limitless possibilities ahead in biology: the final deciphering of the genetic code was in progress, and we thought we knew how genes were controlled. However, the eukaryotic world was largely unexplored, and at the molecular level, we knew nothing about embryology. Never entering my mind at the time was the possibility that I would enter the business arena in a serious way, and it is because I did become involved in business that I am now interested in geology. As I try to reconstruct the events that gave rise to my entry into business, the important elements involve not only what happened in biology that led to biotechnology but, in fact, what happened in the world at large, particularly in the chaotic period of the late 1960s and early 1970s.

I had a wonderful time in Cambridge. I learned genetics from Sydney Brenner and RNA sequencing from Fred Sanger and came home to my first job at University of California, San Diego (UCSD), in La Jolla with the plan of putting those two approaches together to solve important problems in gene regulation. However, in the fall of 1968, when I arrived at the UCSD campus in my tweed coat and rep tie, I was unprepared for what was going on. A demonstration was in progress. The regents and then-Governor Ronald Reagan were meeting at UCSD, and the students were demonstrating in sympathy with the free speech movement going on in Berkeley. As the Vietnam War progressed and our students became increasingly vocal and active in opposition, most of my junior faculty peers and I were led to take part. Within two years, I had shed my tweed coat, and my hair had grown to my shoulders.

Of particular importance to this story was my involvement with a cadre of philosophy students at UCSD in the formation of a free school for our children. The philosophy department was the center of the revolution at UCSD: Herbert Marcuse was there, and one of his students was Angela Davis. Our school, Pepper Canyon, embraced every wacky and fascinating educational fad of the time, from Carl Rogers encounter groups for the parents to Maoism for the students. (We were, of course, kicked off the UCSD campus after one year, but the school actually went on for four years, when the students themselves asked for its dissolution. As far as I can tell, none of them was irreparably harmed by the experience.)

This seemingly irreversible diversion from the world of business actually led to the two most important elements of my involvement with it. First, it was at Pepper Canyon that I met Peter Johnson. Peter was a philosophy student and was one of the founders of the school. Peter was a

good-looking, charismatic, blonde California surfer and probably is one of the smartest people I have ever met, but he became disillusioned with academic philosophy and dropped out of graduate school. When I heard that he was parking cars in a garage in La Jolla, I offered him a job as a technician in my lab, and he quickly picked up not only the technical aspects of molecular biology but its conceptual underpinnings as well. Peter worked in our lab for five years and is a coauthor on some of the papers that Bill Reznikoff and I published in our *lac* operon work (2). Having both been divorced from our first wives in the chaotic Vietnam War years, Peter and I actually shared a house together for a year.

The second important element of my involvement in the revolution is that when gene cloning and the clear opportunities of biotechnology became apparent, I was not inclined to enter into a pact with venture capitalists (nor with my hair down to my shoulders would that likely have happened).

I can remember exactly when I realized what was going to happen in biology. In 1974, Herb Boyer came to UCSD and gave a seminar in which he described how he had cloned a segment of the *Xenopus* rRNA gene into the EcoRI site of a plasmid vector. To me, this was an electrifying event because I had spent years trying to figure out how to maneuver this or that *Escherichia coli* gene to the proximity of a lysogenic phage attachment site so that defective phage particles carrying the gene could be obtained, a kind of genetic cloning. Boyer had discovered a general way to clone any gene from any species, and it was clear that the world had changed.

In this period, my closest friend at UCSD, Mel Simon, and I often discussed the ramifications of gene cloning. The possibilities of what could be done seemed limitless, and the smallness of our labs and the UC bureaucracy too confining. We were well aware of Herb's launch of Genentech and Winston Salser's Amgen venture but felt some disdain for the compromises we imagined they were making, but action seemed imperative, and finally, in 1978, Mel, Joe Kraut (an x-ray crystallographer), Raoul Marquis (a lawyer), and I met in Mel's living room and formed a nonprofit research institute, which we called the Agouron Institute. Agouron was meant to refer to the Agora of Athens, where Socrates met with his friends to discuss ideas, but our spelling is closer to a word meaning "unripe apples" in Greek.

By 1980, we had rented lab space on the beach in La Jolla in the Timken Sturgis building, vacated by the Scripps Clinic in its move to new quarters at Torrey Pines. We began work with a grant from the Navy on a project Mel had proposed concerning bacterial adhesion to surfaces: for the Navy, the marine fouling problem. Our Vietnam era scruples had begun to fade. By 1982, we had about 30 scientists in the Institute and a budget of about $3 million. X-ray crystallographers from Kraut's lab had joined the institute as well as Arnold Hagler's computational group. Former members of my group had learned the intricacies of phosphotriester synthesis of DNA oligomers, and we collaborated with Kraut's group to do the first site-directed changes to be studied at the atomic level (in dihydrofolate reductase) (3).

During this period, we made our first attempt to enter the world of business. Dow Chemical had decided to go into biotechnology and sent an emissary, John Donalds, out into the academic world to make deals. With help from Marquis, we formed a company that we called Syngene and began a long negotiation with Donalds that required many trips to Midland, MI. Syngene would have exploited the opportunities suggested by our early work on protein engineering. In the end, Donalds finally called to make an offer to fund Syngene. When he called, I was actually in the hospital, where I had nearly died after a ruptured gall bladder. The offer was contingent on Marquis having nothing to do with the company. Marquis, an old friend of Mel's, was flamboyant and aggressive in negotiations and apparently had offended Donalds, but Marquis was part of our team, and so I declined Donalds' offer. Shortly after, Mel and I left UCSD and moved to the California Institute of Technology (Caltech).

The Agouron Institute continued, and in some ways, it was already a success. Members at the time recall it nostalgically as a place where there were no grown-ups, but we realized that for it to be a real success, it would have to have an endowment. As professors, we had believed that the University was making a fortune on our overhead, but we came to learn that you are lucky if the overhead actually pays the rent. By this time, Peter Johnson had been out of our lab and doing real estate for about three years, but the real estate market went flat in the early 1980s, and we hired him again, this time to raise money for the Agouron Institute. He quickly became its administrative director and was a success in that role, but the Institute has never received a nickel from philanthropic donors. I once heard Arnold Beckman say that he would never give money to a fly-by-night organization like the Salk Institute. That made it clear what our chances would be. To have an endowment, we would have to earn it ourselves. We did that, and how we did it is part of my story.

In 1984, we decided to start a new company that would capitalize on some interesting things going on in the Insti-

tute. Peter took the initiative in forming the company. He was president of the company during its entire history. This time, we were represented by a bright young attorney from San Diego, Gary Friedman. By this time, a lot of gene cloning companies had been started, and I do not think we would have had success in starting another had our idea not been different. It was based primarily on the work of Dave Matthews, an x-ray crystallographer in Joe Kraut's laboratory. Matthews had solved dozens of dihydrofolate reductase structures bound to a variety of folate inhibitors, such as trimethoprim and methotrexate, obtained mostly from Burroughs Wellcome & Co. From these structures, he felt that he could learn enough to design more potent inhibitors. The idea was to use gene cloning to obtain a drug target, to obtain the crystal structure of the target and of early-stage inhibitor-target complexes, and then to design and synthesize new inhibitors based on that information. In short, we proposed to form a new kind of pharmaceutical company based on the principle of protein structure-based rational drug design. We called it Agouron Pharmaceuticals.

A key aspect in the formation of the company was a commercial cooperation agreement between the Institute and the company in which the Institute would supply technology and would license discoveries to the company. In return, the Institute received the lion's share of the founding stock, 10 times as much as any of the founders, who were Peter, Mel, Gary, Joe Kraut, and myself.

The company got started with small amounts of venture capital, first from a small San Diego company and later from Hambrecht & Quist in San Francisco. Because they were not founders and because their contributions were small, venture capitalists never owned a controlling interest in the company, as they usually do, but instead, in the early years of the company, the Institute was the principal shareholder. We paid for this independence, however, by being chronically undercapitalized. In the end, the growth of the company was primarily fed by public and private stock sales, the first being a small public offering restricted to California, which raised $7.5 million. In all, $200 million was raised in public and private offerings, but we never had more than about $100 million in capital at any time.

The first drug target we picked was thymidylate synthase. Inhibitors of the enzyme are known to be potent agents in chemotherapy, with 5-fluorouracil being the prime example and one still in use. To compete in the market against 5-fluorouracil, a drug would need to be less toxic and perhaps show activity against a different set of tumors. By 1987, the structure of the *E. coli* enzyme had been solved by Matthews, and because its active site was known to be identical to the human enzyme, drug design was begun. To synthesize drugs, you have to have organic chemists, and although there were molecular biologists, x-ray crystallographers, and biochemists in the Institute and the company, there was minimal expertise in organic chemistry. The first organic chemist we hired was a senior pharmaceutical chemist named Terry Jones, who had a deep interest in antifolates. Jones had excellent taste in chemists and hired a terrific group of four or five young Ph.D. chemists from places like Caltech and UC, Irvine. Jones was only in the company for a few years, but his recruits were terrific, and one of them, Mike Varney, eventually became the head of research.

During 1988–1990, a large number of lipophilic antifolate inhibitors of thymidylate synthase in five different classes were synthesized. Many proved to be potent inhibitors of the human enzyme both *in vitro* and *in vivo*, and some of them were active in animal tumor models. Decisions had to be made on which compounds to take forward to clinical trials. Three compounds were chosen, and the first, AG85, was tested for antiproliferative activity in psoriasis. In retrospect, the disease target was a miserable choice, but a success would have been good for the company because there can be a shorter path to Food and Drug Administration (FDA) approval with topically applied drugs. However, the placebo effect in psoriasis is huge, so it is very difficult to demonstrate efficacy, and in fact, we saw none. The stock price, which in small companies is linked to news, dropped.

Two other compounds were selected for trials against cancer, and one of them, AG337, had promising activity in Phase I trials and was taken forward to Phase III trials in chemotherapy against hepatic and head and neck cancer. All of this took a long time and a lot of money, but by 1998, we had to conclude that although AG337 had activity, it was not superior enough to other chemotherapeutic agents to warrant further development. By this time, there was much more on our plate in other areas, and in retrospect, the thymidylate synthase project can be seen as a learning experience in which we developed the tools of rational drug design and transformed the company from a research institute into a small pharmaceutical company that could do toxicology, pharmacology, and pilot-scale synthesis; that could administer clinical trials taking place at diverse sites around the country and in Europe; and that could effectively interact with the FDA.

To do all of this was a tremendous accomplishment that I attribute directly to Peter's growing skill as a manager and a leader. In the early stages of the company, he wore all of the hats. He interacted daily with the scientists and

understood what they were doing; he raised money; he traveled around the country to soothe stockholders and to attract new ones; and he negotiated continuously with the large pharmaceutical companies. As the company grew, a management team that took on many of these responsibilities was formed. The team included Kent Snyder, in Development and Sales from Merrill Pharmaceuticals; Barry Quart, in Regulatory Affairs from Bristol-Myers; Neil Clendenin, in Clinical Affairs from Burroughs Wellcome; and Steve Cowell, the Chief Financial Officer from Amgen. Peter's taste in choosing these people and his tact and skill in motivating them made this group (in fact, the entire company) into a cohesive and congenial organization that could work a miracle. The miracle was a human immunodeficiency virus (HIV) drug called Viracept.

In 1987, with the AIDS epidemic in full swing, Marvin Cassman, a program director in the NIGMS at the National Institutes of Health (NIH; who later became the Director of the Institute), obtained money for a structural biology program to solve key HIV drug targets, and NIH sent out a request for proposals. Although the structure part of this program and its extension into drug design were perfect for us, we had no credentials at all in AIDS research. However, my friend Jim Dahlberg at the University of Wisconsin was a cofounder with Bill Haseltine of Cambridge BioSciences, a company in Boston that specialized in HIV. With Jim's help, a three-way consortium between the two companies and Haseltine's lab at the Dana-Farber Cancer Institute was formed, and we received a $4.5 million grant from NIH. This grant funded study of the structures of three HIV targets: the protease that cleaves viral polyproteins into functional enzymes and structural proteins, the reverse transcriptase (RT), and the integrase. A larger cooperative agreement with Eli Lilly supported work in a number of therapeutic areas, including virology, particularly the drug design portion of this project.

Work was begun simultaneously on the three targets, and by 1991, Dave Matthews had solved the structure of the RNase H portion of RT. On the day that the publication came out, the stock doubled, reflecting the intense perception of a need for an effective HIV therapy in the financial community. RNase H, with its shallow active site, was a difficult drug design target, and our efforts in that direction did not produce any interesting candidates. RT itself was difficult because high-resolution diffracting crystals could not be obtained, and its structure was not determined until much later by Tom Steitz and, independently, Eddie Arnold. Thus, the RT inhibitors came from drug-screening programs in other companies.

We were not the first to solve the structure of the HIV protease. It was solved first by Alex Wlodauer at NIH and then at Merck & Co. We used Wlodauer's coordinates to solve our structure (in a different space group) by molecular replacement. The protease was an ideal drug target, and just about every pharmaceutical company in the world has at one time or another mounted an HIV protease drug discovery program. Our program began in 1991, and within a year, a collaborative team of Agouron and Lilly chemists had progressed from micromolar inhibitors to nanomolar inhibitors. At each stage, crystal structures of drug-enzyme complexes were solved, providing directions for the next generation of compounds. It is well known how to design peptidic transition-state inhibitors of proteases, but peptides are almost never good drugs. To design nonpeptidic inhibitors required determination of the protease ligand structure at each stage. In the end, six molecular classes of protease inhibitor were synthesized. This was the first demonstration, and is still one of the best, of the power of the drug discovery process that had been developed at Agouron during the thymidylate synthase learning period.

Although many of the compounds were excellent inhibitors, a compound called AG1343 was the best candidate, based on its potent antiviral activity. The compound was a product of the joint design effort but was, in fact, synthesized at Lilly, which, accordingly, had the right to its development. If Lilly did not develop the compound in two years, we could exercise our right to develop it.

At this point, a fascinating and, for us, pivotal series of events took place, illustrating the importance of carefully crafted and negotiated contracts as well as just plain luck. In 1992, Lilly had an anti-hepatitis B compound in trials called fialuridine (FIAU). Late into trials and with a delayed hepatic toxicity, a number of patients died. As a result, Lilly decided to exit the antiviral field and terminated the Agouron contract. This meant Lilly would not likely develop AG1343, but at the pace the field was moving, a two-year waiting period for its rights would not have allowed it to be a successful candidate for us.

In its exit from the antiviral field, however, Lilly continued to be interested in upper respiratory infections and thus one of the projects in the joint program: the synthesis of inhibitors of the rhinovirus protease (RVP). Rhinoviruses are the principal cause of the common cold, and a cure for the common cold could be a big product. Quite early in the joint program, Dave Matthews obtained high-resolution diffracting crystals of RVP, but solving the structure was extremely difficult because there were eight molecules per unit cell in a triclinic space group. In the

end, Matthews solved the structure, but it was three months after Lilly had lost its rights to the coordinates.

Peter realized that Lilly wanted the RVP coordinates to continue its own rhinovirus program and dispatched Gary Friedman, our lawyer and negotiator, to Indianapolis to make a deal for AG1343. Friedman returned three days later with complete rights for AG1343 and all of the other compounds that had been developed jointly. In return, we gave them the RVP coordinates. To my knowledge, Lilly never made productive use of the RVP structure.

Thus, one is forced to acknowledge that, for Agouron, AG1343 was as much a business success as an affirmation of the science. AG1343, now known as Viracept, became our blockbuster drug.

The deal for Viracept was made in early 1994, and an ambitious and precarious plan was prepared to bring it to market. Nine months of preclinical trials in 1994 confirmed that Viracept had acceptable qualities. It had little if any toxicity in animals and an acceptable half-life in serum and, most important, was orally bioavailable in several animals. Testing in man was begun in early 1995 in England. In the early Phase I trials on healthy volunteers, it was confirmed that Viracept was orally available and well tolerated. In the following Phase II trials on AIDS patients, the results were initially electrifying. At several different dosage levels, the serum titer of HIV fell more than 2 logs. Concurrent with these drops in viral titer was an increase in CD4 lymphocytes, the primary target of the HIV virus. However, with time, the viral titer in many patients drifted back up due to the accumulation of specific drug-resistant mutations in the protease gene. This had been the earlier experience with RT inhibitors, such as azidothymidine (AZT), and it also was seen with other protease inhibitors. For us, the solution to this problem came in the next series of trials with an observation made on a single patient in San Francisco. In those trials, it was required that the patients be taking only Viracept so that its properties could be ascertained. However, one of the patients in the trial began taking AZT as well as Viracept. In that patient, the virus levels dropped to below detectable levels and remained there. The idea that one should counter resistance by challenging the virus with two drugs makes very good sense, and Merck made the same observations with its protease inhibitor. In addition, it was observed that combining the RT inhibitors AZT and 3TC (2′,3′-dideoxy-3′-thiacytidine) also produced good results. Thus, the best therapeutic outcome is observed when a combination of two RT inhibitors and a protease inhibitor are used in drug therapy. When these results were announced in 1996 at the international AIDS conference in Vancouver, there was a euphoric response in the AIDS community, and that meeting is often considered to be a pivotal moment in the war against AIDS.

For a small company, the demands of mounting late-phase trials were daunting. Not only were there the demands of the trial itself, but one had to make, in carefully graded steps, the assumption that the trial would succeed and to prepare for the market launch of the drug. There is no doubt that we bet the company on Viracept.

In the first place, we spent $150 million on Viracept trials, and we could not have raised that amount of money ourselves. In 1992, we entered into a drug discovery program with Japan Tobacco to discover drugs relating to the immune system, such as immunosuppressants. Japan Tobacco had a monopoly in Japan, but like other tobacco companies, it was trying to diversify and had a fairly large pharmaceutical division. One might question the morality of doing business with a tobacco company, but it had had the good taste not to ask us to join in the therapeutic end of the cancer business, and it was a good business partner. We called it JT. In 1994, JT expanded its Agouron agreement to include antivirals, including HIV. In return for a 50% share in the profits of Viracept, it provided a set of milestone payments eventually totaling $24 million and after that 50% of the costs of further development. In the agreement, Agouron had the rights to market Viracept in North America and JT in Japan. We would jointly decide how to market Viracept in the rest of the world. This part of the agreement was very important to us. No large pharmaceutical company would have given up United States rights completely, and we could not become a real pharmaceutical company without a marketing arm.

Perhaps the most crucial feature of this program was the synthesis of the drug. Viracept is a complicated molecule with five chiral centers whose synthesis requires 20 steps. With time, we built a scale-up facility that could meet the demands of toxicology studies and early-stage trials, but much larger amounts of drug were needed for the Phase II and III trials, and a pipeline had to be built that ultimately would supply the drug for the marketplace. There was finally a need to produce more than 10 metric tons of the drug per month. We could not and did not build a plant to produce that huge quantity but rather farmed out various stages of its synthesis to a number of specialty chemical companies around the world, with the final formulation and packaging taking place at a plant in Puerto Rico. The difficulties in coordinating this complex process, and especially in quality control, cannot be imagined, but to put them into perspective, after their protease inhibitors had been approved, both Merck and Abbott Laboratories

encountered manufacturing problems and for some time had to ration the distribution of drug to pharmacies. Although we many times faced the prospect of such a disaster, Agouron was able to avoid it and continuously decrease the cost of drug synthesis.

Concurrent with the scale-up in the synthesis of the drug was the establishment of a sales force. We could not have built up a sales force to distribute a cardiac drug or an antibiotic, but at that time, there were a manageable number of distribution sites for AIDS therapy, and studies showed that you could handle the United States and Canada with about 100 sales representatives. Kent Snyder built this team in careful stages, with the senior sales managers coming aboard early in the process and the final team being hired with just enough time to be trained before the drug was launched. There are very interesting and complex demands involved in distributing an AIDS drug. The AIDS patient community is organized, intelligent, and vocal. They are capable of understanding a scientific argument, and it is in those terms that the drug must be sold. I met some of our sales representatives, and they were, in general, young and very bright. Some of them were from the gay and minority groups that were most devastated by AIDS, and most of them came from larger pharmaceutical companies.

By the time the pivotal Phase II and III trials of Viracept were commenced in 1995, there were already three other HIV protease inhibitors in trials: Roche Pharmaceuticals' Invirase, Abbott's Norvir, and Merck's Crixivan. The standard was triple-drug therapy, so one could not morally give any patient less than this, and yet for FDA approval, it was necessary to demonstrate efficacy. We designed an innovative trial that met those demands. In a blind trial, patients were given AZT, 3TC, and either a placebo or Viracept. When any patient's viral load went above a certain level, the placebo was switched to Viracept without breaking the blind. The results showed that the viral titer in such patients immediately became undetectable, validating both the efficacy of Viracept and of triple therapy. In the end, 700 patients at 50 different sites in the United States were tested in three different trials. We were also the first to develop a formulation for children and to carry out trials of that formulation. The coordination of the effort and the continuous interaction with the FDA were an immense job directed by our head of Regulatory Affairs, Barry Quart.

Before Christmas in 1996, the entire 2000-pound application was submitted to the FDA 80 days ahead of schedule, and it was approved in February without a formal advisory committee meeting. The whole process, from acquisition of the rights to Viracept from Lilly to approval, took only 35 months, a world record. Toward the end, everyone involved was working 16-hour days and in many cases staying in the office continuously.

Viracept was an immediate success in the market. Although it was the fourth protease inhibitor to be approved, it continuously gained market share and, in 1998, passed Crixivan and became the most prescribed protease inhibitor in the United States, with 30% of the market share. By the end of the first year of sales, 60,000 patients were taking Viracept, and by 1998, there were 90,000 Viracept patients in the United States and another 60,000 in 20 countries worldwide. With this success in the marketplace, Agouron earned its first profit. In 1998, sales of Viracept were $358 million, making its first year the most successful launch in the biotechnology industry.

But to all of us, the most gratifying result of the Viracept story was its impact on the AIDS epidemic. After rising continuously since the beginning of the epidemic, in 1998, the annual number of deaths due to AIDS suddenly dropped by 25%. Overcrowded AIDS clinics emptied. Previously incapacitated people went back to work.

We heard many individual stories, such as this one told by a San Francisco patient (where ddI is dideoxyinosine and d4T is 2′,3′-dideoxy-3′-thiacytidine).

> Life was good. I was successful. I worked hard and built my own company, all set for a comfortable retirement at age 40. Then it started. The strep throats that wouldn't go away, the chills, the blood test that heralds news that you are going to die. How many of us can even accept the idea of death at age 42? Viral load: 86,000. Why is this happening to me? Friends and loved ones lost. My livelihood gone. I put my estate in order; I composed my will. Knowing I was going to die, I said my goodbyes. Other experimental therapies failed; I weakened, and my near-death experience crept ever close. October 1995, I was staring death in the face when I walked into Dr. Conant's clinic. I started my triple combination cocktail: ddI, d4T, and Viracept. The first thing I noticed was I didn't have to nap in the afternoon. Deep within my body the transformation began. I slowly regained strength and energy. Viral load: 500. The best news I've ever heard! In two months complete vitality returns and I live again. Within weeks HIV is non-detectable in my blood plasma—and it's stayed that way since 1995.

In January 1998, Peter sold Agouron to Warner-Lambert, perhaps best known for Listerine and Bubblicious chewing gum but also a large pharmaceutical company with its Parke-Davis subsidiary. The price was $2.1 billion, just half of what Volvo sold for that year. Soon thereafter, Warner-Lambert was sold to Pfizer, and there is still a part of Pfizer research in La Jolla, but the finality of this process is striking. Throughout its history, I was a member of the

Agouron Pharmaceuticals Board of Directors, and I regularly visited the company and talked with the scientists, but after the sale of the company, I never again visited. To me, though, it is pleasing that Agouron Pharmaceuticals had a beginning and an end. This rarely happens in science, so I can still look back with pleasure and pride in what we did.

1) Viracept saved or prolonged the lives of hundreds of thousands of people and continues to do so. However, it must be acknowledged that it was through combination therapy that their deaths were averted, and thus, the credit must be widely distributed between the scientists who characterized the HIV virus, the governments that supported their work, and the other pharmaceutical companies whose drug discovery and distribution programs also contributed. For the surviving AIDS patients in the Western world, at least, the system worked.

2) While acknowledging their contributions, I take pleasure in the fact that we went head to head with two of the biggest and most respected pharmaceutical companies in the world, Hoffmann-La Roche and Merck, and in this race, we won.

3) The enterprise eventually became 1500 people, and together, this team not only derived huge satisfaction and pride from its success but insured the security of their families as well.

4) Many people bought our stock, and I am proud that Agouron Pharmaceuticals was not hype. We actually produced something, and the long-term investors made a lot of money.

But most important to this story, the Agouron Institute was now endowed. We immediately sold the Warner-Lambert stock and invested it in diversified stocks and bonds. The endowment amounted to $80 million, meaning that we could spend about $4 million per year. For several reasons, we decided not to continue with an independent laboratory as we had at the beginning. For one thing, Mel and I were now at Caltech and could see no reason to leave. We did not think that we could make the lab a success if we were not there running it, so we decided to pick specific areas to support, where we could make a difference, and to fund research and education in those areas. We opened an office in Pasadena and hired Joan Kobori as our program director. Joan received her Ph.D. at Stanford with Arthur Kornberg and had worked for a number of years at Caltech with Lee Hood. Nothing that we have subsequently done at the Institute could have been done without her.

The first area we decided to support was supramolecular assemblies: the structure of large functional assemblages in the cell, such as the ribosome, the spliceosome, etc. We held two meetings on this subject in 1998. Out of these meetings came a white paper describing opportunities and needs in this field (see the Agouron Institute Publications web site).

The ribosome was one of the first supramolecular assemblies that could be crystallized, and we gave grants to three of the four groups in this field that eventually solved the structure of the ribosome. This was, in my opinion, one of the great accomplishments of molecular biology. In addition, we gave a grant to the synchrotron facility at the Lawrence Berkeley National Laboratory. Only by use of the synchrotron can crystal structures as large as the ribosome be solved, but many supramolecular structures have proved difficult to crystallize, and we believed that high-resolution cryo-electron microscopy could potentially provide interpretable structures in cases where x-ray crystallography had not succeeded. We bought cryo-electron microscopes for facilities in Caltech, UCSD, and the University of Colorado and gave support for facilities at Cambridge, England, University of California, San Francisco (UCSF), UC, Berkeley, and the Karolinska Institute in Stockholm. Perhaps the most important thing in ensuring the future health of a field is to support training, and we supported 28 structural biologists selected by Jane Coffin Childs or Helen Hay Whitney for postdoctoral fellowships.

Supramolecular structure was a good way for us to start out, but this field is well supported by the NIH, so it is difficult to do something that really makes a difference. Our plan was to stay in an area for five or ten years and then to move on. We wanted to choose a new area in which the relatively small amount that we could spend would make a difference, hopefully an area where we could learn new things.

At Caltech, most of the important academic decisions are made by a committee that consists of the Chairs of the six divisions, the Provost, and the President. There are no academic deans. I was Chair of the Biology Division from 1989 to 1995, and Mel succeeded me, so we knew the other chairs at Caltech. In the Geology and Planetary Sciences Division, the Chairman, Ed Stolper, was faced with an interesting problem. The division had a lot of senior faculty, and so there would be a large turnover. They did a study of where geology was going in the future. They decided that one of the exciting new areas is geobiology. We talked with Ed about this and decided that we would look at geobiology as an area for the Institute to support. For much of the history of the Earth, life consisted mostly of microorganisms, and these microorganisms had pro-

found effects on the geological history of the Earth. In turn, the history of the Earth profoundly influenced evolution. So, in geobiology, it is most relevant to study present-day microorganisms whose metabolic capabilities help to explain what one observes in ancient rocks.

As with supramolecular structures, we began with a meeting and a report. The report (see the Agouron Institute Publications web site) suggested, first and foremost, that the Agouron Institute support an extensive summer course in geobiology. We have done that. The course is organized and given by the University of Southern California at the Wrigley Institute's Marine Science Center on Catalina Island. The course started in 2002 and is still going on. About 20 students from all over the world are selected, and they spend four to six intensive weeks doing geobiology. There is a geology field trip, and the students also study and do microbiology.

The first students in the course have now begun their own labs in geobiology. They were all we could have hoped for. They are field geologists, they are geochemists, and they are microbiologists. They even do molecular biology. They are a small group, they all know each other, and they are fearless. Geobiology at this stage resembles molecular biology in the 1960s. Like we were then, these young geobiologists are the vanguard of a new field.

For the first five years of the course, the field trip was led by John Grotzinger (now at Caltech) and Andy Knoll. I went on all of those field trips, and it is by doing this that I have begun to learn geology. Then, four years ago, John and Andy began a series of advanced field trips that have been three weeks long. On these trips, to Namibia, to Western Australia, and to Oman, we actually did serious geology (Fig. 1).

Another recommendation of our geobiology study was that we undertake a program of core drilling in ancient rock. For many modern geochemical studies, it is necessary to obtain fresh core because the rock on the surface is too weathered. The first such drilling program was in South Africa, and it was to obtain core samples that cover sediments deposited between 2.6 and 2.2 billion years ago. (An entire issue of *Precambrian Research* edited by Andy Knoll and Nick Beukes describing the initial characterization of the Agouron cores has been published (4).) It was during this period that oxygen first appeared in the atmosphere of the Earth. After the origin of life itself, this was arguably the most important event in evolution. To better understand what we were learning from the cores, in 2005, we organized a four-day meeting in Santa Fe, NM, entitled "Oxygen." The participants were biochemists and structural biologists studying photosynthesis and geologists

FIGURE 1. **Ed Stolper, Mel Simon, and John Abelson at the Brockman banded iron formation in Western Australia, 2007.**

who study the Proterozoic Era (2.6 billion to 540 million years ago). Almost all of the field trips I have been on have explored rocks of the Proterozoic Era, and throughout this period, a key question has been the degree to which oxygen in the atmosphere and in the ocean influenced the evolution of life. (I have given several talks on this subject, one of them published (5).) I will briefly summarize this story here because it ties together the field trips, the core drilling, and the meeting and explains why I have come to be so interested in geology.

It has been known for more than 50 years that oxygen first appeared in the atmosphere ~2.3 Ga (2.3 billion years ago). Before that time, the level of oxygen in the atmosphere was $<10^{-5}$ of the current level. In rocks deposited before that time, metals are found mostly in the reduced form; for example, iron is found as pyrite (FeS), siderite (FCO_3), and magnetite (Fe_3O_4). After that time, iron is mostly in red beds, hematite (Fe_2O_3).

The oxygen in the Earth's atmosphere is all generated by photosynthesis. Oxygenic photosynthesis appears to have been invented only once, in cyanobacteria. Eukaryotic algae and plants have colonized cyanobacteria as chloroplasts. A major question is, "When did oxygenic photosynthesis evolve?" Apparently, it evolved by 2.4 Ga, but there is evidence derived from the study of organic biomarkers in rocks that oxygenic photosynthesis may have evolved as early as 2.7 Ga. Roger Summons, an Australian geologist working at the Massachusetts Institute of Technology, has developed sensitive fractionation procedures for the characterization of the organic remains found in rocks as bitumen. In Australian cores from 2.7 Ga, Summons was able to detect steranes. Steranes are the remains of sterols after diagenesis, the alteration of rocks that occurs at high temperatures and pressures. In modern organisms, the biosynthesis of sterols requires molecular

oxygen in 11 separate steps. This is the argument for photosynthesis at 2.7 Ga, but it has been disputed. Summons' techniques are so sensitive that he can detect steranes at parts per billion or even per trillion. Core drilling requires the use of a lubricant, usually oil-based. The drilling equipment must be lubricated, so there is ample opportunity for contamination.

To rule out organic contamination, we drilled the South African cores using only water as the drilling fluid. In addition, the South African cores that document the 2.6 to 2.5 Ga period were drilled in duplicate. One core was drilled in sediments that were at that period deposited near the edge of the continent. The second core site, 20 miles away from the first, is in sediments that were offshore. In both cases, there is about 1400 m of core. Taking multiple precautions to sample only the interior of the core, Summons could prove that, at levels of the core where the sediments at both sites were deposited in deep water, the levels of steranes were identical over 200 m of core (published in Ref. 4). This constitutes the best proof that there was oxygen available before it appeared in the atmosphere, but one wants to know whether this was a local event or over the entire Earth, and also whether oxygen was available for sterol synthesis even earlier. This summer, we roamed all over Western Australia looking at potential drill sites where we can obtain cores of ancient rocks, some as old as 3.5 Ga and some of which contain black shales that may contain biomarkers. (The Australian drilling program is led by Roger Buick, an Australian geologist at the University of Washington. Roger Summons and two other members of the drilling project, Tim Lyons from UC, Riverside, and Ariel Anbar from Arizona State University, were on this trip.) The drilling program will start next year. We will drill about three holes per year for three years.

After the appearance of oxygen in the atmosphere, there was a catastrophic event that could have led to the extinction of all life. The entire Earth froze over, a "snowball Earth" event. I have seen glacial moraines from this period in two places: South Africa and Western Australia. Paleomagnetic measurements show that, in Africa at least, the continent was near the equator in this period. Large drop stones coming from earlier rocks are deposited in fine silt. It is thought that this glaciation may have been caused by the oxidation of methane, a potent greenhouse gas that had warmed the Earth. The snowball Earth event could have lasted for millions of years, but the accumulation of carbon dioxide in the atmosphere from vents eventually provided such a warming effect that the glacier melted.

For the next billion years, not much happened. Roger Buick has called this the "boring billion." There was oxygen in the atmosphere but at only ~10% the present level, and although the surface of the ocean was oxidized, evidence is accumulating that the bottom of the ocean was sulfidic, like the Black Sea is today. This was not an environment that was conducive to multicellular animals, and during this period, cyanobacteria dominated the margins of the oceans.

Our field trips have several times explored the "belt formation" of Western North America. This formation was deposited in a shallow inland sea at ~1.4 Ga, and the fossil remains of cyanobacteria formed in mats. Stromatolites are a prominent fossil feature of these rocks.

The end of the Proterozoic Era was punctuated by two more snowball Earth events: one at 710 Ma (million years ago) and one at 635 Ma. After these glaciations in the Ediacaran Period, oxygen levels in the sea rose, and a strange and wonderful multicellular biota evolved. I have seen fossils from this period in Namibia. They are unlike anything seen since. A fossil called *Swartpuntia* has fronds and is 3-fold symmetric. But late in the Ediacaran Period, an organism called *Kimberella,* discovered in deposits along the White Sea in Russia, appeared. *Kimberella* was a bilateral predator that grazed on the floor of the ocean and resembles modern mollusks. Clearly, the modern gene kit for bilateral development had evolved in the Ediacaran Period. However, at the end of the Ediacaran Period, 542 Ma, there was an extinction event, and all of the Ediacaran fauna disappeared.

This set the stage for the explosion of life forms in the Cambrian Period (542 to 488 Ma). During this period, the continents were arrayed along the equator, and there were abundant sunlit shallow seas. On a field trip to the Bahamas, we flew over this carbonate platform dotted with islands, and it must be the closest one can come to experiencing the Cambrian world. Oxygen in the atmosphere rose to near present levels in the Cambrian period, and the first ancestors of almost all of the present phyla evolved. Multicellular life was on its way.

I am looking forward to next June, when we will go to Spain and study the fossils of the early Cambrian Period.

Acknowledgments—I have known Mel Simon and Peter Johnson for 40 years. The Agouron story is their story, too, and it is Peter's great triumph. It does not get any better than this: to embark on an undertaking with your friends, to succeed, to do good, and to have a lot of fun.

Joan Kobori has been our program director since we had enough money to have a program. The logistics of these endeavors, the meetings, the courses, the projects, the fellowships, etc., are all complicated, but she makes it look easy, and the external impression of the Institute as being informal, unbureaucratic, and friendly is due to Joan. We could not have done this without her.

The Agouron Board of Directors includes Mel (Chairman), Peter, Gary Friedman (still serving as our attorney), Gordon Gill, Debbie Specter, Ted Friedman, Gustaf Arrhenius, Willis Wood, David Hirsh, Ed Stolper, John Grotzinger, and me. The board members have been great. They support our good ideas and dampen out the bad ones. Again, this is about friends having fun. Four of the board members, Gordon, Debbie, Ted, and Gustaf, have been on our board since the beginning. The only way to get off our board is to die, and we have had some losses along the way. Nate Kaplan and Martin Kamen were mentors to Mel and me at UCSD, and they were early members. I miss Nate and Martin a lot and wish that they had lived to see Agouron Pharmaceuticals succeed and to participate in the resulting transformation of the Institute. My uncle, Philip Abelson, was a member since the beginning, and he did live through most of this story. (He died at 91 in 2004.) Phil had been at one time the Director of the Carnegie Institution of Washington Geophysical Laboratory, and he particularly liked the drilling program.

John Grotzinger and Andy Knoll have been my mentors in geology, and judging from what their students have done, I could not have picked better mentors. Andy knows more about geology and biology than anyone since Darwin. He is the quintessential geobiologist. John is widely regarded as the best sedimentologist in the world. In the field, he is amazing. The geologist Paul Hoffman told me that no one else in geology has a sharper eye than John, and after he has spent a short time looking at a formation, he can immediately describe the ancient world at the time those rocks were deposited. I hope I can have many more field trips with John and Andy.

Christine Guthrie is my colleague, my wife, and the love of my life. I could have written a completely different Reflections article on our parallel and complementary life in biochemistry. We, with 10 or 15 of our friends, began the field of RNA processing, and I still work in that field. I have retired from Caltech, and for the past five years, I have been working in Chris' lab at UCSF. At UCSF, I sit out in the lab with the students and attempt to study spliceosome assembly by single-molecule Förster resonance energy transfer. She attends committee meetings, writes grants, teaches, and runs the laboratory. I am pretty lucky to be indulged at UCSF while at the same time free to go off for weeks at a time to some place like Namibia and do geology.

Address correspondence to: johnabelson@gmail.com.

REFERENCES

1. Cohen, P. A., Bradley, A., Knoll, A. H., Grotzinger, J. P., Jensen, S., Abelson, J., Hand, K., Love, G., Metz, J., McLoughlin, N., Meister, P., Shepard, R., Tice, M., and Wilson, J. P. (2009) Tubular compression fossils from the Ediacaran Nama Group, Namibia. *J. Paleontol.* **83,** 110–122
2. Dickson, R. C., Abelson, J., Johnson, P., Reznikoff, W. S., and Barnes, W. M. (1977) Nucleotide sequence changes produced by mutations in the *lac* promoter of *Escherichia coli*. *J. Mol. Biol.* **111,** 65–75
3. Villafranca, J. E., Howell, E. E., Voet, D. H., Strobel, M. S., Ogden, R. C., Abelson, J. N., and Kraut, J. (1983) Directed mutagenesis of dihydrofolate reductase. *Science* **222,** 782–788
4. Knoll, A., and Beukes, N. (eds) (2009) Initial investigations of a Neoarchean shelf margin basin transition (Transvaal Supergroup, South Africa). *Precambrian Res.* **169,** March 2009, Issues 1–4
5. Abelson, J. (2007) The Birth of Oxygen. *Bulletin of the American Academy of Arts and Sciences*, Spring 2007