Photo by: Bob Paz

David Baltimore

ANNUAL REVIEWS

Annual Review of Immunology

Sixty Years of Discovery

David Baltimore

Division of Biology and Biological Engineering, California Institute of Technology, Pasadena, California 91125, USA; email: baltimo@caltech.edu

Annu. Rev. Immunol. 2019. 37:1–17

First published as a Review in Advance on October 31, 2018

The *Annual Review of Immunology* is online at immunol.annualreviews.org

https://doi.org/10.1146/annurev-immunol-042718-041210

Keywords

Mengovirus, poliovirus, reverse transcriptase, Abelson virus, tyrosine protein kinase, NF-κB, microRNAs

Abstract

Each of us is a story. Mine is a story of doing science for 60 years, and I am honored to be asked to tell it. Even though this autobiography was written for the *Annual Review of Immunology*, I have chosen to describe my whole career in science because the segment that was immunology is so intertwined with all else I was doing. This article is an elongation and modification of a talk I gave at my 80th birthday celebration at Caltech on March 23, 2018.

INTRODUCTION

I started my first laboratory at the Salk Institute in the spring of 1965. That means that my laboratory has been doing experimental science for 53 years. I had been in training for 5 years already when I started my own laboratory, so I have been doing science for almost 60 years. That strikes me as miraculous.

Only a few readers can remember back to doing molecular biology in 1960. It was rudimentary. You could do some enzyme biochemistry but you couldn't do much with DNA or RNA. We had no restriction enzymes, no reverse transcriptase, no ligases, no means of sequencing DNA, no vectors; we had none of the tools that produced recombinant DNA techniques some 15 years later. But we did have viruses, and they had bits of DNA or RNA that could be harvested and studied. And that was where I focused when I started out.

As I think back over these many years, the changing concerns of my lab are very evident. The driving force of that changing focus was generally new technical developments, and I will try to make that perspective evident.

ANIMAL VIRUS RESEARCH

I started at MIT as a graduate student in 1960. I first worked on phage, helping out Cyrus Levinthal, a physicist-turned-biologist who was a young faculty member. I worried that phage work was not going to prepare me for the future and asked Cy if one could work on animal viruses rather than bacterial viruses and, with that, start learning about the molecular biology of animal cells. I remember so well Cy's answer: He said, "I have also wondered about that proposition, and I am not sure about the answer."

With that helpful response, I went down the hall to Salvador Luria—a founding father of molecular biology, later to be a Nobel Laureate—and posed the same question. Salva said he too was not sure of the answer, but he arranged for me to find out for myself by spending the following summer working with Phillip Marcus, an animal virologist at Albert Einstein College of Medicine, and taking the then-new Cold Spring Harbor course on animal viruses.

By the end of the summer, I was sure there was a career to be had in animal virology and I decided to do my thesis with one of the directors of the course, Richard Franklin. That involved moving to the Rockefeller University, where Richard was a junior faculty member.

A few words about Salva, as he was universally known: He was an Italian-Jewish immigrant who left Italy, spent some time in France, and was finally drawn to the United States in 1940, where he teamed up with an immigrant from Germany, Max Delbrück. Although they were never at the same university, they worked closely together, meeting in summers at Cold Spring Harbor, searching for the nature of the gene using bacteriophage, and creating the field of molecular biology.

When I met Salva, during a summer I spent at Cold Spring Harbor, he had just moved to MIT, where he and Cy initiated one of the first molecular biology graduate programs in the world. He encouraged me to come to MIT after I finished my undergraduate degree at Swarthmore College, and I gratefully agreed to come. When I said that I wanted to leave after one year at MIT to work with Richard, I was being a bit selfish. Salva, however, unhesitatingly agreed to write a letter of support to the president of Rockefeller, Detlev Bronk, who personally accepted students. That letter got me into Rockefeller, even though it was now August 1961 and I was hoping to start there in September. It was characteristic of Salva that he acted in my interests, not his own. Later he welcomed me back to MIT as a postdoc and later still as a faculty member, each time facilitating my career with funds and position. I consider him my most important mentor, although I never worked with him.

I began working with Richard's pet virus, Mengovirus, a close relative of poliovirus (1). These are RNA viruses, and they pose the particular problem of how RNA is duplicated in an infected cell. That was a problem that my lab worked on for many years thereafter. My initial approach was to use biochemical methods, because that was the cool methodology of the time. The methods I started with were those developed by Arthur Kornberg and his students. I got the ideas from the literature but had to adapt them to the system that interested me. With these tools I could show that these viruses blocked cellular RNA synthesis in the nucleus of the infected cell and set up factories in the cytoplasm for viral RNA production (1). In fact, in my second year as a graduate student, I demonstrated the presence in cytoplasmic extracts of Mengovirus-infected cells of an RNA-dependent RNA polymerase (2, 3), the first ever shown to exist. This defined one type of RNA viruses, the positive-strand RNA viruses. We call them positive strand because the RNA in the virus particles encodes the viral proteins and is therefore infectious: It encodes a polymerase and all other viral proteins. (As an aside, showing how different science was in the early 1960s from what evolved over the next decades, to do my first in vitro experiments I had to synthesize ^{32}P-labeled nucleoside triphosphates.)

Biochemistry, really enzymology, was wonderful and I love it to this day, but it shows what happens in test tubes, not what happens in cells. So I developed a method of probing cells, using an idea that derived from autoradiography, Richard's major tool. But I got rid of the cumbersome process of autoradiography itself and figured out how to get a Geiger counter to do the quantitation. This opened up the study of virus-infected cell physiology and, along with enzymology, was the workhorse of my thesis (4). But as I read and heard about the work of others, I realized that the technique of sucrose gradient centrifugation had been developed and would provide new avenues for study of the questions that intrigued me. (This method is based on creating a stable gradient of sucrose in an ultracentrifuge tube, layering a solution with nucleic acids, proteins, viruses, or organelles on top of the gradient, and submitting the tube to centrifugal forces that then distribute the materials in the layer by mass, allowing the recovery of desired species. Actually, the function of the gradient of changing concentration of sucrose is to stabilize the liquid column through which macromolecules are being pushed by the centrifugal forces, keeping them in a layer as they move through the sucrose.) So, in 1963, I moved for an early postdoc to James Darnell's lab, then at MIT, where this and other techniques were being used (5–7).

I followed Darnell to Albert Einstein College of Medicine in New York City the next year but actually worked there with Jerry Hurwitz, honing my skills in biochemistry (8) and learning from a true master.

TO THE SALK INSTITUTE AND THEN MIT

During the winter of 1964–1965, Renato Dulbecco came to New York City and offered me the chance to have an independent laboratory in his space at the Salk Institute in La Jolla, California. Renato was then a faculty member at Caltech (California Institute of Technology), but he was planning to move to Salk. However, the negotiations of the move went back and forth while I waited to see if the opportunity was really going to materialize. In the early spring, Renato finally said the move was on, and soon I drove across the country to begin at Salk.

I started at Salk continuing to study what poliovirus did to cells that allowed the infected cells to become factories for virus production. Our focus was on viral RNA synthesis. At Salk, Marc Girard was my first postdoc (9) and Alice Huang was the second (10, 11). Work on polio protein production there was initiated by my first graduate student, Michael Jacobson (12, 13). After a couple of years, Salva offered me the opportunity to return to MIT as Associate Professor, and I gratefully accepted. I arrived on the first of January, 1968, and worked there with many postdocs and graduate students for most of the next 30 years.

The most important new tool of that time was SDS polyacrylamide gel electrophoresis. It was a simple way to separate and quantitate individual proteins. Mike Jacobson had started using it at Salk and continued at MIT, ultimately using amino acid analogs to show that poliovirus encoded a single polypeptide representing the entire coding capacity of the virus. That polyprotein was cut up into many smaller proteins, each with a characteristic function. It was a result with important implications for how translation of messenger RNAs in animal cells differed from translation in bacterial cells.

Soon after arriving at MIT, we expanded the laboratory to work on other viruses, notably one that my new wife, Alice Huang, had worked on for her thesis, vesicular stomatitis virus (VSV). We quickly saw that VSV had a radically different mode of infection from poliovirus. Although polio carried the sense strand of RNA—so that it inoculated the cell with a new messenger RNA—VSV, we saw, carried the antisense, or negative, strand in its infectious particles (14). That posed a conundrum, because a negative strand is senseless and cannot start an infectious process. A way out of this quandary would be for the particles to carry a polymerase that could copy the senseless RNA into sensible RNA, using the Watson-Crick base-pairing rules. We assayed for this polymerase, and sure enough it was in the particles (15).

This provided a clear distinction between two types of RNA viruses. Some viruses—the positive-strand viruses—have an infectious RNA in their particles. This RNA can encode the virus's proteins. Their particles contain no polymerase, but they induce production of an RNA-dependent RNA polymerase in infected cells. Other RNA viruses, generally ones surrounded by a lipid envelope, hold a noninfectious RNA but have in their particles a polymerase able to copy this RNA. Their RNA sequence does not encode proteins but is the base pair complement of an encoding strand.

That explained the life cycle of many related enveloped viruses—they are negative-strand viruses like VSV—but one class of viruses was still enigmatic, the RNA tumor viruses. These viruses did not fit the pattern of other viruses. They looked physically like negative-strand viruses, but the RNA in the infecting particles was positive strand. We attacked these viruses biochemically and found their secret. In experiments that I did with my own hands, I found that these viruses had an RNA-dependent DNA polymerase in their particles, implying that their life cycle involved reverse transcription, the copying of RNA into DNA. Howard Temin's laboratory found this enzymatic activity at the same time, and we published back-to-back papers (16, 17). The existence of an enzyme that could synthesize DNA in the virus particles suggested that these viruses might integrate their information into the cellular chromosomes and use their genes to take over the cell. This seemed an important clue about why these viruses cause cancer. All of this turned out to be true. This work extended the central dogma—that DNA can be copied into RNA, which encodes protein—to show that RNA can be copied into DNA. It built on ideas that Howard had been thinking about for 10 years. Howard and I received the Nobel Prize in Physiology or Medicine for this work in 1975, sharing the prize with Renato Dulbecco, whose work showed that DNA cancer-inducing viruses integrate their genes into cellular chromosomes.

HOWARD TEMIN

A few words about Howard Temin and our intertwined lives: I started out in experimental biology because of a chance observation of my mother's. She was an experimental psychologist teaching at the New School for Social Research in New York City. During my junior year in high school, she noticed on a bulletin board a flyer advertising a program for high school students being run at the Jackson Laboratory (Jax Lab) in Bar Harbor, Maine, during the upcoming summer. She told me about it, and I applied and was accepted.

During that summer I, like all of the students, worked with three mentors doing small research projects. I realized later that my mentors were three of the world's great geneticists: Elizabeth Russell, Donald Bailey, and Willys Silvers. My experience with them was so exciting that I decided to structure the rest of my life around becoming an experimental biologist.

The program at Jax had about 27 high school students; one senior college student acted as a sort of guru for us. That was Howard. Howard seemed to know the answer to every question we had. He was in his final year at Swarthmore College. One of the important reasons I decided to go to Swarthmore was because it had educated Howard so brilliantly.

Living near the laboratory in Bar Harbor was a summer resident, a Spanish scientist from New York City named Francisco Duran-Reynals. He believed that viruses were a major cause of human cancer and gave a yearly lecture at Jax about this subject. Few others believed in this causation process, making him something of a heretic. Only later was he proven so very right. It is an amazing fact of history that Howard and I heard this lecture during that summer of 1955, and then 15 years later we provided the mechanism for this process.

Howard went from Swarthmore to Caltech as a graduate student, and it was there that he began to think about how RNA viruses could cause cancer. He went from Caltech to the University of Wisconsin and developed his ideas there. I called him as soon as I realized that I had found the reverse transcriptase, and he told me that his laboratory also had evidence for it. That led us to publish our back-to-back papers.

Everyone who knew Howard understood what a rare and outstanding intellect he was. He died of cancer at the relatively young age of 59, a great loss to cancer research and to the world.

REVERSE TRANSCRIPTION

The finding of reverse transcription opened new directions for me and wrapped up one previous concern. The previous concern went back to my first days as a scientist. I had started working on an RNA virus, and the deep problem that concerned me was understanding its life cycle. I wanted to understand viruses as genetic systems. DNA viruses were easy to understand: They were like tiny cells whose DNA held all of their genetic secrets, and transcription released these secrets to control the behavior of an infected cell. Even single-stranded DNA viruses presented little difficulty: The infected cell could copy the strand into a double strand, and after that transcription would handle matters.

But RNA viruses could not fit this model. The segregation of gene storage in DNA that works so well for DNA viruses would not be easily available with RNA as the genetic material. When I started being interested in this problem, it was known that a drug that prevented DNA-dependent RNA synthesis, actinomycin D, inhibited the infectivity of DNA viruses but did not affect many RNA viruses. Thus, such viruses did not use DNA at all in their life cycle. By searching out polymerases, we showed that there were both positive-strand and negative-strand viruses. The reverse transcriptase finished the picture by showing that there were RNA viruses that used reverse transcription to transfer the sequence of their RNA into DNA, producing a complicated lifestyle of reverse transcription, integration of the DNA into cellular DNA, transcription of the integrated DNA back into RNA, and then packaging of this RNA.

In 1971, I put my thinking about viral genetic systems into a review article (18) that proposed that viruses could be classified into six classes: double-stranded DNA viruses, single-stranded DNA viruses, double-stranded RNA viruses, positive-strand RNA viruses, negative-strand RNA viruses, and retroviruses (those with reverse transcriptases). Virus classification up to that time had used a Linnean system of great complexity. By this simplification, students could easily keep in mind the key aspects of the different types of viruses. It became known as the Baltimore classification and is widely used today.

Experimentally, the discovery of the reverse transcriptase led my laboratory in three new directions. One was largely the work of Inder Verma, a recently arrived postdoc who showed that a viral reverse transcriptase could copy any RNA as long it was provided with a primer DNA (19). Thus, the enzyme could make a cDNA copy of a messenger RNA using as a primer a short polymer of dT residues. Inder's work gave the biotech industry as well as science laboratories a powerful new tool.

The second direction was to understand the details of the reverse transcription process. This work spread out over a decade, leading to a seminal 1979 paper that specified the process in all of its biochemical glory (20). It was the ultimate accomplishment of my focus on biochemistry going back to my earliest days in research.

The third direction was for my laboratory to begin to study cancer. My first experiment on a cancer-causing virus had been to uncover the reverse transcriptase, leading to their name retrovirus. Understanding that viruses might cause cancer by adding genes to cells was exhilarating, and I wanted to take that idea further. But I wanted to work with mice, not chickens, which had been used in much of the earlier work on RNA tumor viruses. So I sought a retrovirus that would be easy to work with, would transform mouse cells in the lab, and was able to rapidly cause cancer in mice. I found the virus a few floors above me at MIT; a postdoc there had discovered such a virus when he was at the National Institutes of Health (NIH), but he was no longer working on it. He was Herbert Abelson, and his virus became known as the Abelson virus. That virus carries a gene we call *abl* that is able to transform normal cells into cancer cells.

We began work on *abl* in 1970 and continued working on this virus for the next 15 years. Naomi Rosenberg did the initial biological work (21, 22) that set the stage for Owen Witte to isolate the Abl protein that caused the cancer, which turned out to have the unique property of being a protein-tyrosine kinase (23). Coincidentally Tony Hunter at the Salk Institute found that this unique biochemistry was associated with cancer induction by other viruses. Twenty years later it was shown that an inhibitor of this biochemical process, a kinase inhibitor called imatinib or Gleevec, acts as a virtual cure for those suffering from a form of leukemia called chronic myelogenous leukemia (CML) (24). CML cells harbor a chromosome fusion that links the *abl* gene to the *bcr* gene and activates the kinase activity of the *abl* gene. George Daley and Rick Van Etten (25) showed that the *bcr-abl* fusion gene caused CML in mice, proving that the fusion was the cause of the cancer. Gleevec was the first kinase inhibitor to be an effective cancer cure and opened a new line of pharmaceutical research, the development of kinase inhibitors as anticancer drugs. It is particularly gratifying to me that work we began in 1970 resulted, 30 years later, in the cure of a cancer in many patients, a remarkable vindication of the belief that basic scientific investigation leads to medical progress in unexpected and certainly unprogrammed ways.

One other consequence of the discovery of the reverse transcriptase was administrative within MIT. Salva was then, in 1970, a recent recipient of the Nobel Prize in Physiology or Medicine, a recognized father of the field of molecular biology, and a man who had followed my career very closely. He decided that my foray into cancer research had such wide-ranging implications that MIT should open a major effort in cancer research. President Nixon's War on Cancer had just been announced, and it included establishment of cancer centers. Most would be significantly clinical in orientation, but MIT had no medical school. We decided to start a center studying the basic science of cancer. Luria got the agreement of Jerry Wiesner, the MIT president, wrote an application to the government for the funding of such a center, and was awarded funds for the only nonclinical cancer center to be part of the War on Cancer. We found that MIT owned a building then being used to make chocolates for Brighams, a venerable Cambridge confectionary and ice cream store. I helped Luria transform the building into a research center, which opened

its doors in 1974, just in time to be a site for celebrating my 1975 Nobel Prize for discovering the reverse transcriptase.

I was 37 years old when I received the Nobel Prize. I had already been in research for 14 years, had been a full professor for a number of years, and felt myself something of an elder. I mention this because today the training time for a research scientist has become so prolonged that the average first-time recipient of an NIH grant is 42 years old (mine came when I was 28). I believe that we must give independence to biomedical scientists at a much younger age if we are to fully nurture the creativity that is inherent in newly emerging investigators.

MOVING INTO IMMUNOLOGY

When I was at Salk in the 1960s, there had been two big laboratories devoted to immunology, one run by Ed Lennox and the other by Mel Cohn. The trainees in those labs loved to talk about immunology, and they introduced me to the challenging questions about how the immune system works. Those questions intrigued me, but in the 1960s I could not see how to attack them and stayed with viruses.

Then, in the 1970s, three things happened that opened the opportunity for my laboratory to work in immunology. First, Susumu Tonegawa—in a tour de force, using very primitive methods—showed that nucleic acid recombination was at the heart of the diversity of immune function (26). Second, recombinant DNA methods came together, allowing investigators to cut, paste, and duplicate DNA at will. It was pretty obvious that the recombinant DNA methods were the right tools to blow open immunology.

Third, a clinician, Ronald McCaffrey, joined my laboratory and wanted to search for evidence of a retrovirus as the cause for human leukemia or lymphoma. The most direct assay for such a virus was to look for a reverse transcriptase in tumor tissue, and numerous investigators had undertaken such searches in a variety of cancers with tantalizing suggestions of the presence of the enzyme but no definitive evidence. Ron ground up lymphoma tissue and assayed it. I distinctly remember a night when he called me and with great excitement said that he had positive evidence of reverse transcriptase in some samples. My job as mentor was to calm him down and make sure that he had done all the necessary controls. He had not, but the next day he set up the experiment with the requisite controls, and sure enough, it was not reverse transcriptase. However, it was another DNA polymerase—called terminal transferase (TdT)—that he was assaying, an enzyme that made DNA but did not copy a template. It was an enzyme known to be in thymus tissue, suggesting that the positive tumors might arise from thymus. Ron was desolate that he had not found a virus—and only in the case of one rare tumor have investigators definitively detected such a virus in human tumors to this day—but I was intrigued that such an enzyme should be specifically found in the immature immune cells of the thymus. In 1974, I published a short theoretical paper suggesting how the enzyme might help generate diversity in antibody genes (27). It was a rare theoretical paper for *Nature* to publish and proved to open the field into a productive direction.

I decided in 1976 that my laboratory should become involved in the study of immune cell differentiation. Luckily a few of my postdocs, notably Fred Alt, Enzo Enea, and Al Bothwell, were as intrigued as I was by the puzzles of immunity, and they got the lab into experimental immunology. By 1980 we had developed the skill and understanding to begin to pose and answer questions about the details of immunoglobulin gene rearrangement and expression (28, 29).

I note above that my postdocs got the laboratory into immunology. I phrased it that way because about this time I stopped doing experiments myself, and all the work of the lab was done by trainees. This is a common progression of professors who reach a point when they no longer

can find the block of free time needed to carry through an experiment. I actually did experiments myself for much longer than professors do today, many of them leaving the bench when they set up their first independent laboratory. This is an understandable but deplorable trend because it takes from laboratory experimental work the person with the greatest skill and experience. Actually, I stopped doing experiments for another reason. It occurred on a particular day in about 1976. That day I said to myself, "If we are going to start doing immunology, I should set a good example and do some of the work myself." The first experiments we planned involved recombinant DNA methods, methods which had just been regulated by the NIH through a process established at the Asilomar meeting I had helped to organize. These regulations specified that work had to be done using a safe bacterial strain, pBR322, that could not grow outside the laboratory. So my first work was to set up a culture of pBR322. I did this, and the bacteria grew slowly, as was expected for this highly genetically compromised strain. I then transferred the bugs to fresh medium, and now they grew much more rapidly. I knew that that was a bad sign, indicating that I had contaminated the culture or selected a mutational variant. Al, who was already very skilled with pBR322, took pity on me and said I should retire from the laboratory and leave the experiments to him. This I did, and since then I have not done an experiment.

Somewhat earlier, our interest in immunology got a boost from another part of the laboratory when we realized that bone marrow cells transformed into tumor cells by the Abelson virus were held at the differentiation stage of immature B lymphocytes and that individual clones of cells were arrested at different stages in the B cell differentiation pathway (30). Our assay for differentiation state was the rearrangement of the DNA at the immunoglobulin loci. We found that some clones were rearranged at the heavy chain locus and some at both heavy and light chain loci but never was one rearranged solely at the light chain locus. This provided evidence for an ordered pattern of rearrangement: first heavy chain, then light chain (31, 32). In fact, in some clones not all the cells were rearranged; these represented clones that were undergoing rearrangement. This gave us a biological system to study rearrangement (33).

THE RAG GENES AND DNA REARRANGEMENT

Later, we sought the genes that encoded proteins that catalyzed the process of immunoglobulin gene rearrangement. This involved a lengthy series of experiments initiated by David Schatz, one of my students. He was joined by another student, Marjorie (Margie) Oettinger, in the landmark cloning of the recombination-activating genes (RAG genes). The first experiments, done by David, were to demonstrate that human DNA encoding the RAG activity could be transferred into mouse fibroblasts. This was accomplished by making 3T3 fibroblasts that contained a gene construct encoding a selectable marker that could be activated by the immunoglobulin-gene-rearrangement machinery (34). The experiment showed that although the 3T3 cells have the RAG genes, because they contain the complete genome, the genes are inactive. However, DNA taken from cells that have the inactive genes can be broken in such a way that the genes become active when transfected into 3T3 cells.

Once the genes were transferred into the 3T3 cells, the next task was to purify them. This monumental task was accomplished in two steps, because it turned out that there were two genes (RAG-1 and RAG-2) (35, 36). Luckily, those two genes were close to one another in the DNA; otherwise they would have been separated during the transfer to 3T3 cells and their activity would have been lost, because both gene products are required for immunoglobulin gene rearrangement. Once DNA containing the individual genes was obtained, it was easy to show that recombination required both of them. The two students who purified the genes have continued to study their action in their own laboratories at Yale (David) and Harvard (Margie). As well, many other

investigators have devoted themselves to understanding RAG-mediated recombination, notably Martin Gellert at NIH. However, there remain many enigmatic aspects to their activity.

One of my favorite conundrums in immunology has been allelic exclusion. It comes about in the following way. An early observation—and a foundational aspect of the clonal selection hypothesis of antibody production—was that any one immune B cell makes only one type of antibody: one heavy chain and one light chain. But every B cell, being diploid, has two heavy chain loci, multiple light chain loci, and multiple T cell receptor (TCR) loci. How does a cell know to stop rearrangement after successfully rearranging only one of its heavy chain loci and then one of its light chain loci? This was the problem of allelic exclusion, and Fred Alt and I spent long hours trying to understand its molecular basis. We never completely solved this problem, although we saw how the problem might be solved. Fred has continued to study that key question for many years, and it is gratifying that the explanations we imagined turned out to be right.

IMMUNOLOGY AS DEVELOPMENTAL BIOLOGY

To me, the formation of the many different cell types that make up the immune system is a classic problem in developmental biology. We focused our attention on the B cell, the cell that makes antibodies, for a systematic analysis of key questions about its development. We were helped in this work by our discovery that the Abelson virus immortalizes immature B cells, making intermediates in B cell differentiation available for detailed study. But the key work was done by Cary Queen, a postdoc, who showed that when a κ light chain gene was transfected into a myeloma cell it was transcribed from a promoter element but that transcription depended on a downstream element we later defined more precisely and showed had the properties of an enhancer (37, 38). This was one of the earliest demonstrations of the role of enhancers in the transcription of mammalian genes. Cary also observed that the enhancer activity was evident in lymphoid cells but not fibroblasts, showing the tissue specificity of the enhancer.

We as well as other labs spent much effort defining the regulatory sequence elements that led to tissue-specific transcription of immunoglobulin light and heavy chain genes. The fact that transcription was tissue specific implied that immune cells contained proteins that activated transcription. It seemed likely that such proteins bound to the regulatory sequences, so a postdoc of mine, Ranjan Sen, teamed up with a postdoc in Phil Sharp's lab, Harinder Singh, to develop an assay for protein binding to specific DNA sequences. They modified an assay in the literature to come up with a binding assay called the electrophoretic mobility shift assay (EMSA), because it detected binding by a shift of mobility of a piece of DNA in an electric field stabilized in a polyacrylamide gel (39). Ranjan then broke the known regulatory DNA sequences into small pieces and found many proteins detected by EMSA that bound to individual pieces of the DNA (40). One of these seemed strikingly tissue specific and bound to a site in the light chain enhancer. We called it NF-κB because it was a nuclear factor that bound to the κ enhancer in B lymphoid cells. Other proteins detected by EMSA bound to one or another regulatory region and were picked up for study by other members of my laboratory. One such protein bound to a highly conserved DNA octamer in the promoter of immunoglobulin genes and has been the subject of much study (41), including the cloning of the gene for the binding protein in my laboratory by Lou Staudt (42). A multitude of productive investigators coming from my lab and many other labs have devoted their attention to understanding the roles of the proteins uncovered by EMSA. Their work has made developmental immunology a very exciting and productive field. But I chose to focus much of the work in my laboratory on the one factor we had discovered that turned out to have little developmental significance.

NF-κB

My own laboratory focused its attention on the NF-κB protein, ultimately showing that it is not tissue-specific but rather exists in all cells. However, in most resting cells it is bound to an inhibitory protein (IκB) that masks it from DNA binding (43, 44). In B lymphoid cells, IκB is constitutively degraded so that NF-κB is free of masking and registers in EMSA. Thus its tissue-specificity is a consequence of IκB being active in all nonlymphoid cells. However, NF-κB can be released in all cells by treating them with proinflammatory proteins (TNFα, LPS, IL1, etc.). Exposure to these inducers activates degradation of IκB by the ubiquitin/proteasome system. NF-κB then activates transcription of hundreds of genes. These observations established NF-κB as a key—arguably *the* key—mediator of inflammation in the body. Inflammation being one of the most important protective responses of the body—fighting off possible pathogens and other noxious invasions, as well as being in its chronic form a contributor to many diseases—makes NF-κB one of the most interesting and important transcription factors in cells.

Having expected to find developmentally significant factors, we tested NF-κB for a developmental role by knocking out, or obtaining a knock out, of the genes that encode the various proteins that make up the NF-κB family of proteins. Interestingly, animals lacking these proteins, singly or even in pairs, develop normally. There is only one developmental problem. Lack of one protein, the RelA protein or p65, does cause neonatal lethality but this effect, late in development, is due to TNF-induced liver toxicity that is normally countered by NF-κB and is therefore not a true developmental problem.

A number of generations of the people who have studied with me have taken on aspects of the NF-κB story, continuing to the present day, and the rewards have been rich. Furthermore, many other laboratories have studied aspects of NF-κB function. We have learned that NF-κB is a family of 15 dimeric proteins drawn from a pool of 5 monomers. There are numerous secondary modifications of the NF-κB monomers, which have regulatory roles. There are many review articles and even books (45) that chronicle the remarkable story of how this small family of proteins can control so many downstream proteins and have effects on a multitude of disease states. Most recently, an important role for NF-κB in cancer has been extensively documented.

HIV

Another line of work we started later in the 1980s was to understand the workings of HIV, the virus that causes AIDS. We began this work because the virus was so intriguingly different from any other virus in having a reverse transcriptase but being a lytic virus, not a cancer-inducing one. I also had cochaired a panel that proposed the United States have a billion-dollar research program on HIV/AIDS, and in the report of the panel we encouraged scientists working on other problems to take an interest in understanding HIV. I felt that I had to take my own advice. That program is also continuing in my laboratory to this day.

Remarkably, early on in our work on HIV, Gary Nabel, an MD/PhD postdoc, realized that the helper T cells that grew the virus had active NF-κB. He showed that the virus has NF-κB binding sites in its transcriptional promoter, explaining a key aspect of the virus's life cycle (46). His work showed just why HIV only grew in cells that had been activated and linked our work on NF-κB and its proinflammatory activation to our work on HIV.

Above, I have described four lines of work my laboratory undertook. I started with virology, moved into cancer, became an immunologist, and then incorporated study of HIV. In taking on these multiple lines of work, we were always cognizant of new technologies that could be applied to the problems that interested us. Some of the new technologies we perfected, like the mobility shift assay, and some we took from the literature. One key method was the genetic alteration of

mice. When it first became feasible to delete or insert a gene in a mouse, we were an early adopter because the addition or removal of a gene was a great method for probing the role of particular genes in immune functions. We did a few deletions; others did many and made it a routine aspect of investigations of immune function.

TRANSGENE ACTIVITY AND THE US CONGRESS

One of the earliest genetic alterations we did in mice was to implant a rearranged, functional immunoglobulin heavy chain transgene into the mouse genome. This had the effect of suppressing rearrangement of endogenous heavy chain genes (47) and strengthened the understanding that allelic exclusion was a feedback process mediated by functional immunoglobulin protein.

The transgene also caused a strange alteration in the repertoire of rearrangements that did occur. Our analysis of this phenomenon was done as a collaboration with Thereza Imanishi-Kari, an immunologist with many years of experience studying immune repertoires who had been a colleague at MIT and had moved to Tufts University (48). A postdoc in her laboratory believed that there had been scientific misconduct in the course of this work and thus started a 10-year controversy that even involved the US Congress. It is all described in a book written by Daniel Kevles (49), and I will not go into much detail here. The upshot of the controversy was that Thereza was exonerated, but because I defended her over those 10 years, I was accused of misconduct in the press and even brought in front of a congressional committee, where I verbally sparred with the notorious Congressman John Dingell. I was also the object of opposition by scientist colleagues who wanted the controversy to disappear at whatever cost to me. I attempted to keep my laboratory functional through this time and the major effect on me was that I was forced to cut short my presidency of the Rockefeller University—discussed in the next section—as I fought the negative publicity surrounding the situation.

ADMINISTERING SCIENCE

Having mentioned one of my administrative positions, let me detour from talking about science and discuss my administrative roles. Starting in 1980 and lasting to 2006, I took on various roles in the administration of institutions of science. I did this while continuing to run an active laboratory, mentoring trainees, and publishing extensively.

By 1980, my laboratory was working on poliovirus and cancer and had recently taken up immunology. One day I received out of the blue a call from a wealthy entrepreneur named Jack Whitehead. He had amassed a fortune from having created an automated clinical analysis method that was used worldwide. He wanted to use $35 million of his resources to build and equip a laboratory building that would house a basic research institute, which he would endow with a further $100 million. These numbers look small in today's dollars, and when compared to the billions of dollars being invested in research today by Mark Zuckerberg and others. However, in 1980 this was big money and Jack was one of the very rare people to propose investing private money in basic science on that scale. His philanthropy can only be compared to what John D. Rockefeller and Andrew Carnegie invested in basic science in the early twentieth century. Jack's call to me was a request that I come to New York to meet with him and his very impressive board of advisors—including Joshua Lederberg, the president of the Rockefeller University and Gus Nossal, from the Hall Institute in Australia—to advise them on the possible focus of a Whitehead Institute. I arrived in New York having given the opportunity some thought and suggested to the group that they consider a focus on developmental biology, an area then coming into prominence.

They liked the proposal so well that the next day I received a call from Jack, who proposed that I lead the effort to create the Whitehead Institute. This meant finding a site—assumed to be in

the penumbra of an existing strong institution—constructing a building, finding faculty members, and creating a world-class institution. The administrative challenges of that process cut into the time I could spend on my lab, but I had assembled a group of stellar people and they carried forward our program. We built the institute in affiliation with MIT, my home institution for the previous 12 years, and that made it easier to both continue my science and take on the building process. This occupied me from 1980 to 1989, and by that time Whitehead was recognized as a world-class research institute.

The development of the Whitehead Institute was not without its difficulties. MIT had never taken on the role of harboring an independent research institute, and many faculty members were concerned that the wealth of the institute, its $100 million endowment for fewer than 20 faculty members, would allow it to dominate hiring in biology and would skew MIT's program in biology toward directions that the other biology faculty might not have wanted to take on its own. There was also concern, however unfounded in reality, that Jack might influence the directions of the institute and that he might seek to profit from it. The *Boston Globe* highlighted the concerns in its coverage of the birth of the Whitehead Institute, exacerbating the situation. I tried hard to keep my head and to ameliorate the concerns by meeting with the various factions. The MIT faculty meeting at which a vote was taken on accepting the association with a Whitehead Institute was the largest gathering of MIT faculty since the Vietnam War. The vote was positive, although by no means overwhelmingly so. As I left the meeting I called Jack to tell him it was a go, and he left me with the memorable comment that democracy is a wonderful process if you win.

In 1990, I left MIT and the Whitehead Institute to return as President of my alma mater, the Rockefeller University. It was a great institution, but it was founded in 1901 and had not renewed its structure over the 90 years since. I hoped to spend the next decade overseeing that process. I thought that the Thereza Imanishi-Kari affair was settled and would not interfere with my life anymore, but it returned with such force that I could not continue as Rockefeller's president, resigning Thanksgiving of 1991. I stayed on as a faculty member for a couple of years and then returned to MIT, becoming a university professor.

When I moved to Rockefeller, I maintained my laboratory but curtailed the scope of its activities. When I ended my administrative activities, I expanded that scope again. We worked on all of the various questions that interested me. I had become fascinated by the roles of small, compact motifs in proteins, typified by the Src homology regions, SH2 and SH3, found in tyrosine-specific protein kinases, specifically the ones associated with the *abl* oncogene. Today, we are quite used to the idea that conserved domains found in many proteins carry out stereotyped binding reactions, but when Tony Parsons and Hidesaburo Hanafusa recognized that the class of tyrosine kinases had conserved noncatalytic protein segments, the role of such domains was obscure. Mutagenesis showed that they played a role in oncogenesis by the tyrosine kinases. Bruce Mayer, a postdoctoral fellow with me, showed that the SH2 domain bound to phosphotyrosine (50), the product of the kinase activity, as others had suggested. Also, we put significant effort into understanding the specificity of the SH3 domain, finding a proline-rich target for binding by SH3 (51).

I came back to MIT in 1994 and continued to work in cancer and immunology. In June 1996, a government legal panel issued a complete exoneration of Thereza, saying that the Office of Research Integrity (ORI), which had handled the investigation, had failed to prove even one of its 19 charges. The panel "found that much of what ORI presented was irrelevant, had limited probative value, was internally inconsistent, lacked reliability or foundation, was not credible or not corroborated, or was based on unwarranted assumptions" (52).

I expected that to be the end of my peripatetic and administrative tendencies but very soon was encouraged to put my name in for the presidency of Caltech. It seemed to me a long shot, having never spent more than two days at Caltech. I later found that all Caltech presidents had been

new to the campus except Lee Dubridge, who spent a little time as a postdoc there. Following an exhaustive vetting—after all, I had been the object of federal investigation and stood up against a powerful congressman, never mind having been opposed by important elders in my own field—I was asked to take the job by the Board of Trustees. With great excitement and anticipation, I accepted. Caltech is a rare organization, a hybrid of a university and a research institute. Caltech has a contract to manage the Jet Propulsion Laboratory (JPL), an activity mainly supported by NASA, which handles most US interplanetary exploration. JPL has a budget three times the rest of Caltech and is an important part of Caltech, with its own director. With great excitement over the new challenges presented by Caltech/JPL, I accepted the presidency and started there in October 1997. To this day, I remain a professor at Caltech.

Moving to Caltech, I seriously considered closing my lab but luckily did not. However, I swore that I would not take any students while I was president and would take only a few postdocs. One graduate student who joined the Division of Biology a few years after I came to Caltech, Lili Yang, insisted that I was the only faculty member she wanted to work with, and I relented. That was a propitious decision, because Lili set a new direction for the lab.

TRANSLATIONAL SCIENCE

Lili had wanted to study the TCRs on regulatory T cells. She set out to make a lentivirus vector into which she could clone the genes for the two chains of the TCR. She did that, and the vector worked incredibly well (53)—so well that we wondered if this could allow us to reprogram the specificity of human T cells as a mode of cancer therapy. To test this idea we teamed up with Toni Ribas at UCLA, and after a few years we were treating melanoma patients with cells containing transduced TCRs that recognized a tumor antigen called MART-1 (54). The method worked but not well enough to provide cures, and Toni is continuing the approach by transducing hematopoietic stem cells.

Lili's success with a TCR-containing lentivirus vector suggested that there might be other ways that vectors could enhance the abilities of immune cells. To test this general notion, we committed the laboratory to a translational research program we call Engineering Immunity. We applied for support for this to the Gates Foundation, which funded us generously with a Grand Challenge Grant.

With the Gates funds in hand, we set out on four programs of investigation and expanded the lab to bring in leaders for the various program elements. Each program ultimately generated one or more papers describing ways that vectors could be used. Three of the programs are in commercial development. Lili acted as the general manager of the overall effort. It has spun off two start-up companies. One of these, Calimmune, used small interfering RNAs expressed from a lentiviral vector to inhibit production of a cellular protein, CCR-5, needed for HIV entry into cells (55). This company was bought by a larger pharmaceutical company that continues exploring ideas that were developed in Calimmune.

The other company we formed is now a public company called Immune Design. It is exploiting an idea generated by Lili and Pin Wang, a professor from the University of Southern California, of making a dendritic cell–specific lentiviral vector (56). This vector delivers DNA into dendritic cells and produces a very powerful stimulus to antigen-specific T killer cells. The vector is therefore a very effective tumor vaccine; it has been shown to stimulate human antitumor T cells against the antigen NY-ESO-1 and is entering phase 3 clinical trials (57).

A third approach developed as part of the Engineering Immunity program uses an adeno-associated virus vector expressing genes for a monoclonal antibody as a prophylactic vaccine against HIV (58) and is now in clinical testing at NIH. This was the brainchild of a postdoc, Alex

Balasz. It is potentially a very powerful mode of delivery of monoclonal antibodies with activity against infectious agents of many sorts, like malaria (59) or influenza virus (60).

microRNAs AND SPLICING CONTROL

Along with our translational work, soon after coming to Caltech my laboratory began to work on microRNAs, a program of research that continues to this day. microRNAs are short RNAs that are involved in gene regulation in very subtle ways. They were only discovered in the 1990s, and we are still developing our understanding of the many roles they play. I thought they were likely to be involved in regulating the immune system. Our effort was initiated by two postdocs, Mark Boldin and Konstantin Taganov (61). The program has been carried forward by many other trainees, notably a group of MD/PhD students in a joint Caltech/UCLA program.

We are also working presently on splicing control of gene expression following an inflammatory stimulus. We first found that the ordered expression of genes following such a stimulus was not controlled transcriptionally but rather by splicing (62). We then showed, in work not yet published, that delayed splicing was caused by retained introns. Most recently Luke Frankiw, an MD/PhD student who is destined to be my last graduate student, has shown in work under review that a protein involved in splicing control acts on retained introns to facilitate their splicing.

THE END IS NIGH

The group who are presently in my laboratory are the last trainees I expect to have. I have been involved in research for almost 60 years, and I think it is time to leave the field to younger people. The way research is done today, the way it is reported in the literature, the time it takes trainees to pass through their stages of training, the volume of literature produced yearly, all of this is very different from when I entered experimental biology. Meanwhile, like any 80-year-old, I am going physically downhill. I am glad to say that half of my lab space is to be taken over by a new assistant professor in the fall of 2018. This progression is as it should be.

Can I draw any lessons from the 60-year string of experiments that I have overseen? One thing is that laboratories are sites of dual activities: generation of knowledge and training of the next generation of scientists. As a supervisor, I have felt the responsibility to support both perspectives, letting my trainees take the credit they are due for their fantastic work and, as important, letting postdocs take their projects on into their independent careers. For me that created an opportunity to change the focus of the lab as new people arrived. That is the secret behind the ever-changing scientific challenges we have undertaken. Each new project derived from previous ones but posed new questions, thrust us into new directions. Leading the lab to constantly attack new questions has been exhilarating for me because the science has never gotten stale. It has forced me to adapt new techniques as they were invented, to learn the context of new fields and to interact with new communities. I could never have done it this way if I hadn't been lucky enough get a string of the finest nascent scientists to join the laboratory and grasp the opening opportunities ahead of them. That I could lead this extraordinary group of people in this enterprise has been the gift of a lifetime, and I wouldn't have wanted it any other way. I made the decision to devote my life to basic biomedical science before I went to college, and I never wavered from that commitment.

DISCLOSURE STATEMENT

The author is not aware of any affiliations, memberships, funding, or financial holdings that might be perceived as affecting the objectivity of this review.

LITERATURE CITED

1. Franklin RM, Baltimore D. 1962. Patterns of macromolecular synthesis in normal and virus-infected mammalian cells. *Cold Spring Harb. Symp. Quant. Biol.* 27:175–98
2. Baltimore D, Franklin R. 1962. Preliminary data on a virus-specific enzyme system responsible for the synthesis of viral RNA. *Biochem. Biophys. Res. Commun.* 9:388–92
3. Baltimore D, Franklin R. 1963. Properties of the Mengovirus and poliovirus RNA polymerases. *Cold Spring Harb. Symp. Quant. Biol.* 28:105–8
4. Baltimore D. 1964. *The Diversion of Macromolecular Synthesis in L-Cells Towards Ends Dictated by Mengovirus*. New York: Rockefeller Inst.
5. Baltimore D, Becker Y, Darnell JE. 1964. Virus-specific double-stranded RNA in poliovirus-infected cells. *Science* 143:1034–36
6. Darnell J, Penman S, Baltimore D. 1965. Molecular events in the synthesis of poliovirus. In *Perspectives in Virology IV*, Vol. 16. ed. M Pollard, pp. 16–33. New York: Harper Row
7. Baltimore D, Girard M, Darnell J. 1966. Aspects of the synthesis of poliovirus RNA and the formation of virus particles. *Virology* 29:179–89
8. Maitra U, Novogrodsky A, Baltimore D, Hurwitz J. 1965. The identification of nucleoside triphosphate ends on RNA formed in the RNA polymerase reaction. *Biochem. Biophys. Res. Commun.* 18:801–11
9. Baltimore D, Girard M. 1966. An intermediate in the synthesis of poliovirus RNA. *PNAS* 56:741–48
10. Baltimore D, Huang AS. 1970. Interaction of HeLa cell proteins with RNA. *J. Mol. Biol.* 47:263–73
11. Huang AS, Baltimore D. 1970. Initiation of polyribosome formation in poliovirus-infected HeLa cells. *J. Mol. Biol.* 47:275–91
12. Jacobson MF, Baltimore D. 1968. Polypeptide cleavages in the formation of poliovirus proteins. *PNAS* 61:77–84
13. Baltimore D, Jacobson MF, Asso J, Huang AS. 1969. The formation of poliovirus proteins. *Cold Spring Harb. Symp. Quant. Biol.* 34:741–46
14. Stampfer M, Baltimore D, Huang AS. 1969. Ribonucleic acid synthesis of vesicular stomatitis virus. I. Species of ribonucleic acid found in Chinese hamster ovary cells infected with plaque-forming and defective particles. *J. Virology* 4:154–61
15. Baltimore D, Huang AS, Stampfer M. 1970. Ribonucleic acid synthesis of vesicular stomatitis virus. II. An RNA polymerase in the virion. *PNAS* 66:572–76
16. Baltimore D. 1970. Viral RNA-dependent DNA polymerase. *Nature* 226:1209–11
17. Temin HM, Mizutani S. 1970. RNA-dependent DNA polymerase in virions of Rous sarcoma virus. *Nature* 226:1211–13
18. Baltimore D. 1971. Expression of animal virus genomes. *Bacteriol. Rev.* 35:235–41
19. Verma IM, Temple GF, Fan H, Baltimore D. 1972. In vitro synthesis of DNA complementary to rabbit reticulocyte 10S RNA. *Nat. New Biol.* 235:163–67
20. Gilboa E, Mitra SW, Goff S, Baltimore D. 1979. A detailed model of reverse transcription and tests of crucial aspects. *Cell* 18:93–100
21. Rosenberg N, Baltimore D, Scher CD. 1975. In vitro transformation of lymphoid cells by Abelson murine leukemia virus. *PNAS* 72:1932–36
22. Rosenberg N, Baltimore D. 1976. A quantitative assay for transformation of bone marrow cells by Abelson murine leukemia virus. *J. Exp. Med.* 143:1453–63
23. Witte ON, Dasgupta A, Baltimore D. 1980. Abelson murine leukemia virus protein is phosphorylated in vitro to form phosphotyrosine. *Nature* 283:826–31
24. Druker BJ, Lydon NB. 2000. Lessons learned from the development of an Abl tyrosine kinase inhibitor for chronic myelogenous leukemia. *J. Clin. Investig.* 105(1):3–7
25. Daley GQ, Van Etten RA, Baltimore D. 1990. Induction of chronic myelogenous leukemia in mice by the $P210^{bcr/abl}$ gene of the Philadelphia chromosome. *Science* 247:824–30
26. Tonegawa S, Hozumi N. 1976. Evidence for somatic rearrangement of immunoglobulin genes coding for variable and constant regions. *PNAS* 73(10):3628–32
27. Baltimore D. 1974. Is terminal deoxynucleotidyl transferase a somatic mutagen in lymphocytes? *Nature* 248:409–11

28. Alt FW, Bothwell ALM, Knapp M, Siden E, Mather E, et al. 1980. Synthesis of secreted and membrane-bound immunoglobulin μ heavy chains is directed by mRNAs that differ at their 3′-ends. *Cell* 20:293–301
29. Alt FW, Enea V, Bothwell ALM, Baltimore D. 1980. Activity of multiple light chain genes in murine myeloma cells producing a single, functional light chain. *Cell* 21:1–12
30. Siden EJ, Baltimore D, Clark D, Rosenberg NE. 1979. Immunoglobulin synthesis by lymphoid cells transformed in vitro by Abelson murine leukemia virus. *Cell* 16:389–96
31. Siden E, Alt FW, Shinefeld L, Sato V, Baltimore D. 1981. Synthesis of immunoglobulin μ chain gene products precedes synthesis of light chains during B-lymphocyte development. *PNAS* 78:1823–27
32. Alt F, Rosenberg N, Lewis S, Thomas E, Baltimore D. 1981. Organization and reorganization of immunoglobulin genes in A-MuLV-transformed cells: rearrangement of heavy but not light chain genes. *Cell* 27:381–90
33. Lewis S, Rosenberg N, Alt F, Baltimore D. 1982. Continuing kappa gene rearrangement in a cell line transformed by Abelson murine leukemia virus. *Cell* 30:807–16
34. Schatz DG, Baltimore D. 1988. Stable expression of immunoglobulin gene V(D)J recombinase activity by gene transfer into 3T3 fibroblasts. *Cell* 53:107–15
35. Schatz DG, Oettinger MA, Baltimore D. 1989. The V(D)J recombination activating gene, RAG-1. *Cell* 59:1035–48
36. Oettinger MA, Schatz DG, Gorka C, Baltimore D. 1990. RAG-1 and RAG-2, adjacent genes that synergistically activate V(D)J recombination. *Science* 248:1517–23
37. Queen C, Baltimore D. 1983. Immunoglobulin gene transcription is activated by downstream sequence elements. *Cell* 33:741–48
38. Bergman Y, Rice D, Grosschedl R, Baltimore D. 1984. Two regulatory elements for immunoglobulin κ light chain gene expression. *PNAS* 81:7041–45
39. Singh H, Sen R, Baltimore D, Sharp PA. 1986. A nuclear factor that binds to a conserved sequence motif in transcriptional control elements of immunoglobulin genes. *Nature* 319:154–58
40. Sen R, Baltimore D. 1986. Multiple nuclear factors interact with the immunoglobulin enhancer sequences. *Cell* 46:705–16
41. Staudt LM, Singh H, Sen R, Wirth T, Sharp PA, Baltimore D. 1986. A lymphoid-specific protein binding to the octamer motif of immunoglobulin genes. *Nature* 323:640–43
42. Baltimore D. 2011. NF-κB is 25. *Nat. Immunol.* 12:683–85
43. Baeuerle PA, Baltimore D. 1988. Activation of DNA-binding activity in an apparently cytoplasmic precursor of the NF-κB transcription factor. *Cell* 53:211–17
44. Baeuerle PA, Baltimore D. 1988. IκB: a specific inhibitor of the NF-κB transcription factor. *Science* 242:540–46
45. Karin M, Staudt LM. 2010. *NF-κB: A Network Hub Controlling Immunity, Inflammation, and Cancer*. New York: Cold Spring Harb. Lab. Press
46. Nabel G, Baltimore D. 1987. An inducible transcription factor activates expression of human immunodeficiency virus in T cells. *Nature* 326:711–13
47. Weaver D, Costantini F, Imanishi-Kari T, Baltimore D. 1985. A transgenic immunoglobulin μ gene prevents rearrangement of endogenous genes. *Cell* 42:117–27
48. Weaver D, Albanese C, Costantini F, Baltimore D. 1991. Retraction: Altered repertoire of endogenous immunoglobulin gene expression in transgenic mice containing a rearranged mu heavy chain gene. *Cell* 45:247–59
49. Kevles D. 1998. *The Baltimore Case: A Trial of Politics, Science and Character*. New York: Norton
50. Mayer BJ, Jackson PK, Van Etten RA, Baltimore D. 1992. Point mutations in the *abl* SH2 domain coordinately impair phosphotyrosine binding in vitro and transforming activity in vivo. *Mol. Cell. Biol.* 12:609–18
51. Ren R, Mayer BJ, Cichetti P, Baltimore D. 1993. Identification of a ten-amino acid proline-rich SH3 binding site. *Science* 259:1157–61
52. Res. Integr. Adjudic. Panel. 1996. *Thereza Imanishi-Kari, Ph.D.* DAB 1582, Jun 21, Dep. Health Hum. Serv.

53. Yang L, Qin X-F, Baltimore D, Van Parijs L. 2002. Generation of functional antigen-specific T cells in defined genetic backgrounds by retrovirus-mediated expression of TCR cDNAs in hematopoietic precursor cells. *PNAS* 99(9):6204–9
54. Chodon T, Comin-Anduix B, Chmielowski B, Koya RC, Wu Z, et al. 2014. Adoptive transfer of MART-1 T-cell receptor transgenic lymphocytes and dendritic cell vaccination in patients with metastatic melanoma. *Clin. Cancer Res.* 20(9):2457–65
55. Qin X-F, An DS, Chen ISY, Baltimore D. 2002. Inhibiting HIV-1 infection in human T cells by lentiviral-mediated delivery of small interfering RNA against CCR5. *PNAS* 100:183–88
56. Yang L, Yang H, Rideout K, Cho T, Joo KI, et al. 2008. Engineered lentivector targeting of dendritic cells for in vivo immunization. *Nat. Biotechnol.* 26:326–34
57. Pollack SM, Lu H, Gnjatic S, Somaiah N, O'Malley RB, et al. 2017. First-in-human treatment with a dendritic cell-targeting lentiviral vector-expressing NY-ESO-1, LV305, induces deep, durable response in refractory metastatic synovial sarcoma patient. *J. Immunol.* 40:302–6
58. Balazs AB, Chen J, Hong CM, Rao DS, Yang L, Baltimore D. 2012. Antibody-based protection against HIV infection by vectored immunoprophylaxis. *Nature* 481:81–84
59. Deal C, Balazs AB, Espinosa DA, Zavala F, Baltimore D, Ketner G. 2014. Vectored antibody gene delivery protects against *Plasmodium falciparum* sporozoite challenge in mice. *PNAS* 111:12528–32
60. Balazs AB, Bloom JD, Hong CM, Rao DS, Baltimore D. 2013. Broad protection against influenza infection by vectored immunoprophylaxis in mice. *Nat. Biotechnol.* 31:647–52
61. Taganov K, Boldin M, Chang KJ, Baltimore D. 2006. NF-κB-dependent induction of microRNA miR-146, an inhibitor targeted to signaling proteins of innate immune responses. *PNAS* 103:12481–86
62. Hao S, Baltimore D. 2013. RNA splicing regulates the temporal order of TNF-induced gene expression. *PNAS* 110:11934–39