Perspectives

Anecdotal, Historical And Critical Commentaries on Genetics

Edited by James F. Crow and William F. Dove

Recollections of HOWARD TEMIN (1934–1994)

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IN 1960 HOWARD TEMIN finished a postdoctoral year at Caltech and moved to the University of Wisconsin. His brilliance was evident to his Caltech associates, but as to promise he got mixed reviews. GEORGE BEADLE and RAY OWEN both recognized HOWARD's ability but differed in their assessment of his future. RAY was confident, BEETS not so sure. They decided to bet a fifth of whiskey on whether HOWARD would do something important within five years of graduation. After five years he had a publication record that was respectable but not unusual, and he had kept plugging away at an heretical idea. RAY was particularly impressed by one of HOWARD's papers and thought his accumulated work good enough to win the bet, but BEETS demurred. A few years later he reluctantly conceded and the bet was paid off (a bottle of Old Crow, we are told). Of course, if they had waited until the discovery of reverse transcriptase in 1970, there would have been no question.

HOWARD was honest, strongly individual, and willing to state and stand by his convictions. These traits were evidenced at his graduation from Swarthmore, where he had had a disagreement with a college policy and refused to dress for the graduation ceremony. (Years later, after he became famous, Swarthmore awarded him an honorary degree; this time he donned cap and gown.) His outspokenness was also evident at the 1975 Nobel Prize banquet in Stockholm, when he shocked the audience by expressing his outrage that the one known major cause of cancer, cigarette smoking, was so blithely overlooked, even at the event itself. At a gathering of Nobel laureates in 1991, one speaker said that if the world were more loving, many social problems would be solved and world tensions lessened. HOWARD, with AIDS in mind, used his turn to shock an international television audience by saying, "If you love, use a condom." Perhaps the most characteristic example was his steady belief in and defense of the provirus theory during a time when the transfer of information from RNA to DNA was unthinkable.

HOWARD MARTIN TEMIN was born on December 10, 1934, in Philadelphia. He was bracketed by two broth-

ers, MICHAEL (now an attorney) and PETER (now a professor of economic history). His father HENRY was an attorney, his mother ANNETTE (née LEHMANN) was what we might now call a volunteer. Both were strong-minded and voluble. HENRY conducted a diverse one-man legal practice; at home he held high expectations of those around him. ANNETTE had wanted to be a physicist but found that world closed to women. A bright and busy person, she founded a Citizens' Committee for Public Education in Philadelphia and a free summer camp for children who would otherwise be denied that experience. She was active in both Hadasah and the synagogue, where she chaired the School Committee. Thus, whatever may have been the genetic contributions to his abilities, HOWARD also benefited from an environment in which both analysis and action reigned.

As an undergraduate at Swarthmore, HOWARD studied biology. He quickly became a practicing scientist, participating in two summers of research at the Jackson Laboratory. His first paper, dealing with the genetics of congenital anomalies (INGALLS *et al.* 1953), was based on work done there. His final oral examination in 1955 for an honors degree at Swarthmore is well remembered at that institution, because his thoughtful and far-seeing responses elicited a vigorous debate among the external examining committee that soon subverted the examination into a professorial exchange.

There have been numerous tributes to and reviews of HOWARD's life in science (*e.g.*, COOPER *et al.* 1995). Here, instead, are personal accounts by two of us who knew him well, but at different times and in different ways. The first deals with his virology and his Caltech years, the second with his life in Wisconsin.

Caltech and virology (J.W.D.): HOWARD initiated his Caltech graduate work in 1955. He settled first into the laboratory of ALBERT TYLER, a kindly, scholarly developmental biologist who was interested in fertilization and early development. I had been working in the same lab, both of us having arrived at Caltech with an interest in embryology. We were impatient, however, and saw no

FIGURE 1.—HOWARD in 1977 with his favorite mode of transportation in Madison.

way to advance the field, the probes of the time often being no better understood than their target tissues.

After a while, as word of our frustrations spread, JIM WATSON intervened. He was a visiting scientist at the time, fresh off DNA and an object of considerable local curiosity. He decided that we might profitably work with RENATO DULBECCO, a virologist who had recently invented plaque assays for animal viruses and was about to apply the attack mode of the Phage School to these previously intractable viruses. DULBECCO agreed that we could work in his lab and introduced us to the members of his group who were to become our daily mentors. These included MARGUERITE VOGT, who was in charge of overall operations and was intimately familiar with the technical procedures involved in the plaque assays. Another was HARRY RUBIN, a senior research fellow, an adventuresome thinker, and a singer and guitar player much in demand at parties.

Probably because of perceived parallels between developmental processes and the cellular modifications wrought by tumor viruses, HOWARD took up Rous Sarcoma Virus (RSV), which had been discovered by PEYTON ROUS in 1911 and was the canonical “cancer virus.” HOWARD spent most of his subsequent career

exploring this and related retroviruses. At Caltech he promptly engaged in a vigorous collaboration with HARRY RUBIN. Their first success was the development of an RSV assay suggested by the then-recent results of MANAKER and GROUPÉ (1956). This “focus” assay was a hybrid between a virus-induced plaque and a cell colony, in which RSV-transformed cells locally out-proliferate their contact-inhibited siblings (TEMIN and RUBIN 1958). Just as with the invention of the phage and the animal virus plaque assays, the focus assay brought all the analytical power of quantitation to bear on the question of virus-induced cell transformation.

1950–1959 was a decade of great excitement in biology. There was much discussion at Caltech about the still mysterious prophage state in lysogenic bacteria. By the middle 1950s it had become clear that the prophage was so closely associated with the bacterial chromosome that it behaved like a bacterial marker. Thus, HOWARD’s mind was soon steeped in the powerful concept of the chromosomal prophage, and he began to imagine “lysogeny” by an RSV “provirus.” He later found an RSV variant that produced a variant morphology in transformed chicken cells, further strengthening the analogy with bacterial lysogeny.

In 1960 HOWARD moved to the University of Wisconsin, where he remained for the rest of his life. He continued to probe the nature of the RSV-transformed cell and began to advocate in print the notion that a provirus state could be achieved by an RNA tumor virus (*e.g.*, TEMIN 1963, 1964a). He surmised that the RNA of the virus would have to be copied into the DNA of the provirus and that progeny virus could result from transcription of this provirus. However, in addition to assuming a different shape and becoming tumorigenic, most or all RSV-transformed cells continuously release virus without dying. This was very different from the prophage → phage transition so elegantly elucidated by ANDRÉ LWOFF a few years earlier in which an occasional cell suddenly bursts, releasing many phage particles.

In support of his hypothesis, HOWARD found that blocking transcription with actinomycin D stopped virus production (TEMIN 1963), whereas transformed cells continued to make virus in the presence of inhibitors of DNA replication such as fluorodeoxyuridine, aminopterin, and cytosine arabinoside (TEMIN 1964b). Then, using hybridization techniques, he demonstrated sequence homology between RNA from the virus and DNA from transformed cells (TEMIN 1964c). However, these studies are more persuasive in retrospect than they were when first described, in part because of complexities in some of the results and universal uncertainties in the interpretation of inhibitor studies, but probably more because of the novelty of the hypothesis.

HOWARD’s prophage model required that viral single-stranded RNA be copied into double-stranded DNA and that the latter be integrated into the host DNA. Early attempts to find an enzymatic activity to catalyze

FIGURE 2.—HOWARD TEMIN and SEWALL WRIGHT in an animated discussion in 1980.

these steps in infected cells produced only negative results. Only several years later did he find the requisite activity, reverse transcriptase, which turned out to be sequestered in the virus particles themselves. The enzyme was simultaneously discovered by DAVID BALTIMORE (BALTIMORE 1970; TEMIN and MIZUTANI 1970). For this discovery, TEMIN and BALTIMORE received the 1975 Nobel Prize in Physiology or Medicine (along with DULBECCO for his pioneering work with virulent animal viruses).

The power of the provirus hypothesis and its implied biochemistry was attractive to some of HOWARD's associates. However, the notion was less compelling to most virologists, and the walk to consensus was very much uphill. This is a period for which close scrutiny might contribute interestingly to the history of science, particularly to the problem of how scientists come to accept a radical notion. Much was made of the apparent contradiction between the provirus hypothesis and CRICK's "Central Dogma" of the unidirectional flow of information from DNA (through RNA) to protein. The streak of antireligious humor that turned a paradigm into a dogma was unfortunate, for CRICK had later to clarify that his dictum did not preclude information flow from RNA to DNA but stressed instead the impossibility of information flow from protein to nucleic acid. An attractive but far less explicit alternative hypothesis for HOWARD's results, and one under vigorous examination in those days, was that of a "carrier" state, an ill-defined association between viruses and infected cells. Work on the carrier state produced a bin full of arcane phenomenology that could be interpreted in many ways without embracing the TEMIN hypothesis. Although then still

undiscovered, RNA plasmids might have offered an even more compelling (albeit incorrect) hypothesis to explain the carrier state. By standing firm, HOWARD later gained praise for his courage in the face of widespread disbelief of his thesis.

HOWARD did a great deal of virology during his decades in Madison, publishing some 276 research and review articles on the subject in addition to his Caltech output and training several-score graduate students and postdoctoral fellows. Starting even before Madison, he repeatedly wrote on the role of viruses in cancer, a vexing question before the discovery of oncogenes, and he argued for the origins of retroviruses from cellular transposons.

HOWARD became steadily more interested in the problem of viral variation. Even in the Caltech years it had been obvious that RNA viruses mutate rapidly, frustrating attempts to obtain stable genetic markers. From the middle 1970s onwards he worked on this problem. He showed that reverse transcriptases exhibit high error rates (MIZUTANI and TEMIN 1976) and that viral mutations not only occur at high rates (ZARLING and TEMIN 1976; DOUGHERTY and TEMIN 1986) but are sometimes bizarre (PATHAK and TEMIN 1990a,b). He also showed that the two retrovirus genomes that are packaged together in the typical particle recombine at a high rate (HU and TEMIN 1990a,b), further enhancing viral variability. This high rate of variation complicates both the prevention and the treatment of RNA-virus infections.

The Wisconsin years (J.F.C.): I first noticed HOWARD in seminars. Upon his arrival in the fall of 1960, he immediately started his life-long custom of sampling widely in several departments. His breadth of knowledge and depth of insight were apparent in the frequent questions he asked, always to the point. At about the same time there began an active discussion group devoted to peace. Most of those attending were motivated to control nuclear weapons and prevent war, but HOWARD stood out. He had a detailed technical knowledge of arms control, weapons, and military strategies. He more than any other kept the group from degenerating into vacuous do-goodism.

My next notice of HOWARD, some months later, was the increasing attention he was paying to my graduate student, RAYLA GREENBERG. They met in 1961, RAYLA having returned from a year spent in Edinburgh with CHARLOTTE AUERBACH. RAYLA and HOWARD married in May of 1962. I didn't lose RAYLA; I gained HOWARD.

HOWARD was recognized for his knowledge and intelligence. Yet, his provirus hypothesis was received with skepticism, if not downright hostility. His move up the academic ranks was not rapid: four years as assistant professor, five as associate. His promotion to full professor came only a year before his discovery of reverse transcriptase in 1970 made him instantly famous. While serving as Acting Dean, I obtained some unexpected

funds and used them to increase the salaries of a few people who seemed particularly promising. One was HOWARD, but my action led to a complaint from HAROLD RUSCH, then Chairman of the Department of Oncology. RUSCH had a detailed schedule of salaries for his faculty and my move had thrown the plan out of balance.

Scientifically, the years from 1960 to 1970 were frustrating ones for HOWARD. Although he was eager to explain his ideas to anyone who was able to understand them, and did this with vigor and patience, he was generally greeted with disbelief. MICHAEL BISHOP (1995) describes listening in on a long discussion between HOWARD and the skeptics at a Gordon Conference in 1968. The argument was "strong, even vitriolic." HOWARD met all the arguments with reasoned answers, but his was the lone voice of support among the many dissenters, and he failed to convince.

In 1964 HOWARD gave me a manuscript reporting that virus-infected cells contained DNA that was complementary to RNA from the virus, whereas uninfected cells did not. This seemed to HOWARD like the evidence he was seeking and I communicated it to the PNAS (TEMIN 1964c). I was innocent of any detailed technical knowledge, but greatly respected HOWARD's judgment. The paper was widely regarded as unconvincing. Hybridization techniques then were not what they are now, and the presence of other retroviral genomes, unsuspected at the time, interfered. But HOWARD steadfastly continued to view this paper as an important part of his argument.

Is there a lesson to be learned from this? HOWARD's critics found each experiment wanting in some important respect, yet the several separate lines of evidence all pointed in the same direction. HOWARD accepted this (DULBECCO 1995), but the novelty of his hypothesis caused many to continue to doubt. In much of biology, there is rarely a single definitive experimental result. Evidence grows from a number of sources, none perhaps singly conclusive, but impressive in their cumulative impact. In retrospect, HOWARD was right. I wonder how much farther along the biology of retroviruses would be now if his work had gained acceptance in 1964 rather than only after reverse transcriptase was demonstrated chemically six years later. Would our ideas of an RNA basis of early life, our understanding of retrotransposons, our concepts of HIV activity, and our biotechnology be six years farther along?

In any case, the definitive evidence came in 1970. In that summer I was in Japan. HOWARD mailed me a copy of his paper along with a letter of thanks for my confidence in him when the theory was in great disrepute. He was finally vindicated. It was characteristic of HOWARD to take the trouble to write a letter of thanks.

HOWARD became instantly famous. His standing on the Wisconsin campus changed accordingly, and when the Nobel Prize came in 1975 nobody was surprised.

He began to publish more, partly because of a plethora of writing invitations. Nevertheless, paper after paper was full of new data. In the decade of the 1960s he published 27 papers; by contrast, in the 1970s there were 93 and in the 1980s, 104.

He immediately set to work to discover whether reverse transcriptase (which he preferred to call RNA-dependent DNA polymerase) was a normal cell constituent and found that indeed it was. RNA transposons were not only present, but abundantly so. Although he contributed to numerous symposium volumes, he kept up his laboratory work and with his students produced a steady stream of important papers. In addition, he became even more involved in political and social affairs. He was eager to lower the town-gown barrier, taking time to talk with legislators and inviting the Governor to his lab. Yet, he was always in active contact with his laboratory and was involved in the day-to-day work. Increasingly he was drawn into the AIDS question, and one of his last papers presented a novel strategy for developing a vaccine (TEMIN 1993).

On the Wisconsin campus, HOWARD played an increasingly influential role right up to the time of his death. When DONNA SHALALA came to the University as Chancellor, she relied heavily on HOWARD's judgment about University policies, especially research policies. This kind of behind-the-scenes, thoughtful, informal advice was HOWARD's style. His influence was felt in many ways, and no doubt wiser decisions emerged from the Chancellor's office thanks to his input. When SHALALA moved to Washington as Secretary of Health, Education and Welfare, she continued to rely on HOWARD for advice. It was on his suggestion that she invited HAROLD VARMUS to become Director of the National Institutes of Health, an exemplary choice. In Washington as a member of the National Cancer Advisory Board, HOWARD was a strong advocate of investigator-initiated research. He was an outspoken opponent of the Vietnam war, using an occasion during the Stockholm festivities to make his point publicly. He opposed too much expansion of cancer research, citing other needs with higher priority and promise. He got involved in trying to define and cope with scientific dishonesty. I remember a lawn party in which he conducted a poll, systematically asking each of his guests for an opinion.

Despite his heavy schedule of research and the innumerable demands on his time, HOWARD was somehow able to take all this in his stride. He liked to bicycle to work and did so when the Madison weather permitted; when it didn't, he walked. He took great interest in his garden and flowers, and kept detailed, computerized records. He met with his students every Friday afternoon at a conference known as the "confession session." He avoided having too many students and post-docs and was always acquainted in detail with what each was doing. He met for lunch Monday and Tuesday each week with like-minded colleagues for an always-stimulat-

ing discussion of science and public affairs. HOWARD liked to teach and felt it his responsibility to do so. For more than 30 years he taught in the basic virology course that he had organized. Despite national and international commitments, he was in Madison and easily available most of the time. A call to his phone was usually answered by him personally.

HOWARD's Jewish heritage meant a great deal to him, and he regularly attended Sabbath services. Always an avid reader, he had an extensive library of Judaism. He loved reading the Torah and often had a copy with him. He enjoyed discussions with his Rabbi and was especially interested in Jewish history and archeology.

HOWARD had a strong institutional loyalty. He naturally received offers of other positions, but he preferred to stay in Madison. Part of the reason was that he and his family liked their home and their life. HOWARD was close to his wife, RAYLA, and their two daughters, SARAH and MIRIAM. He spent many hours with them, and their interests took highest priority in his schedule. He was respectful of his daughters' feminism and political activism, and joined them in symbolically expressing sympathy for AIDS victims at a White House ceremony; the occasion was HOWARD's receipt of the National Medal of Science in 1992.

HOWARD liked to travel, and after each trip his friends were invited to his home to hear about it and see the numerous colored slides that he brought back. A trip for HOWARD was often a working trip, reading books in advance and giving seminars and visiting labs while there. After touring China in 1977 as part of a trade mission, he wrote a report on the state of biological research (TEMIN 1978). Most memorable was a trip to Russia in 1976. He took with him several forbidden items, including Hebrew books and recent scientific journals. Before leaving, he obtained lists of scientists, especially refuseniks and dissidents. He prepared for the trip by learning the Cyrillic alphabet and bringing a pocket flashlight; these enabled him to find apartments at night. In this way he was able surreptitiously to meet dissidents and refuseniks and to attend their clandestine "Sunday Seminar." Notably, he obtained and brought back a recording taped at the apartment of ANDREI SAKHAROV along with a message to the outside world from SAKHAROV himself. These evening expeditions were made in considerable fear, both for himself and those he was visiting. The distressing details of the life of dissident Russian scientists are spelled out in a diary of his visit (TEMIN 1977). HOWARD brought back valuable information for such groups as Amnesty International. The Madison evening when he told his guests about the trip was especially poignant for one attendee, my newly acquired graduate student, ALEXANDER GIMELFARB, recently a refusenik himself.

HOWARD's final illness, premature, unexpected, and ironic, was a body blow to the Wisconsin community. This man, Wisconsin's best known scientist, the indis-

pensable faculty member, was stricken by the very disease that he had devoted his life to. To add to the irony, the cancer started in the lung, although HOWARD had been a lifelong nonsmoker; however, his was an adenocarcinoma, not associated with smoking. His treatments, many of them experimental, were devastating. But he was determined and stoic. To the extent humanly possible—superhumanly, it seemed—he carried on with research conferences, writing, and advising policymakers. He published 15 papers in the short period, a little over a year, between his diagnosis and his death. HOWARD had always been a voracious reader, and in his last days he read more than ever, especially biographies. One of my last contacts with HOWARD was lending him a biography of R. A. FISHER. His last public talk was in October, 1993 at a Chicago symposium celebrating the fortieth anniversary of the WATSON-CRICK model of DNA (TEMIN 1995). His death came on February 9, 1994 at the age of 59.

In planning the remainder of his life, HOWARD assisted in organizing a 60th birthday symposium featuring his former students and postdocs. Alas, he did not live to attend the symposium, which was held in October, 1994. The resulting published book includes several of HOWARD's key papers along with contributions from former students and associates, a fitting memorial (COOPER *et al.* 1995).

HOWARD is survived by his wife RAYLA GREENBERG TEMIN and two daughters, SARAH and MIRIAM. RAYLA carried out several studies on the concealed viability and sterility loads in natural populations of *Drosophila*. More recently, she has turned her attention to the *Segregation Distortion* phenomenon and, among other things, demonstrated the independent distorting activity of the *Enhancer locus*, *E(SD)*. She is an unusually conscientious teacher and regularly offers an introductory genetics course; she is held in great admiration and affection by her students.

We thank RAYLA TEMIN and BILL SUGDEN for carefully reading an early draft and offering thoughtful suggestions. RAYLA provided documents and shared personal memories that have enriched this article. We thank RAY OWEN for his recollections and MATT MESELSON and FRANK STAHL for their critical comments.

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