Journal of BUON **15:** 174-181, 2010

SPECIAL ARTICLE

Examining the discovery of the human retrovirus

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Summary

Retroviruses have been found in many bird and animal species where they often cause various types of cancer. Dr. Robert Gallo's contribution to the field of retrovirology and the link he established between RNA viruses and cancer has been significant. Historical aspects of his discoveries in the area of human retroviruses are presented and an attempt is made to focus attention on his outstanding role.

Key words: AIDS, HIV, retrovirus

Introduction

In his early years at NIH, Dr. Robert C. Gallo focused his experiments on the comparative biochemistry between active molecular components outside the nucleus of both normal and leukemic human blood cells. His aim was to unravel key differences between the two cell systems which might shed some light on the mechanisms of leukemia induction [1]. Later, he became convinced that nothing exciting would come out of all the comparative biochemistry, in so far that there was no way to distinguish between culpable biochemical changes inducing cancer, and secondary changes playing absolutely no role in cancer induction. He thus started looking elsewhere for fresh ideas and new leads [1].

In the early 1970s, Howard Temin hypothesized that all RNA tumor viruses transcribe their RNA genome into DNA (proviral DNA) which they insert into the genome of the cells they infect. Within a year, Temin and David Baltimore discovered an enzyme, named reverse transcriptase, which mediates the transcription of viral RNA into proviral DNA [2]. In 1975, these two scientists shared the Nobel Prize in Medicine for this discovery. Expectedly then, their achievements during this time period, proved catalytic in shaping Gallo's thinking and in redirecting his work since the tools of molecular biology had been refined [3].

Viruses are of two kinds: DNA and RNA. DNA viruses are those whose genetic information is encoded in DNA format, while RNA viruses are those whose genetic information is encoded in RNA format. Special RNA viruses, called Retroviruses, can convert their RNA into DNA upon infection. This DNA (the provirus) then integrates into the DNA of the host cell. Viruses can take over the metabolic machinery of cells to serve their own purpose. Once in control of cells, viruses seek to replicate themselves (e.g. the flu viruses), change the functional character of their host cells (e.g. some tumor viruses) or do both (e.g. infectious tumor viruses). Rare forms of DNA viruses can integrate their genetic material directly into the DNA genome of cells. Among RNA viruses, only retroviruses have this potential and only after they transcribe their genetic RNA material into the intermediate DNA form (the provirus). This transcription process occurs inside the cell following viral invasion and is mediated by reverse transcriptase, which the retrovirus carries along. Once integrated into the genome of a target cell, the provirus becomes a permanent component of that cell and its progeny.

Induction of cancer by viruses in various animal species had by then been firmly established by several investigators: from poultry to mammals and, on to primates in the wild, but in man. Additionally, the genetic core of the first known cancer virus in animals, the Chicken Rous

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Received 10-07-2009; Accepted 13-08-2009

Sarcoma Virus, was successfully isolated intact. The activation of oncogenes (cancer-causing genes of unknown origin at the time) from an inert state by radiation, chemicals, chance mutations, and other viruses, was theorized by some as a hypothesis explaining the origin of all tumors in all species. We now know that retroviruses sometimes captured some of these genes and made them part of their own genetic information. It was the discovery of the mechanism of reverse transcription by Howard Temin and independently by David Baltimore, that opened wide the field of Molecular Retrovirology, though still limited to animal retroviruses [1]. These investigators succeeded in demonstrating and explaining the conversion of RNA viruses into a DNA form. This DNA form was named Provirus by Howard Temin. Moreover, the discovery of reverse transcriptase [2], confirmed an earlier hypothesis by Temin: that the life cycle of a retrovirus includes an intermediate DNA form. Soon, that integration of infectious, proviral DNA into the genome of target cells and the subsequent role of the same as retroviral oncogenes, was confirmed by many groups. More important, infectious retroviruses were being found in many animal species where they often cause cancers, especially leukemia and other disorders of blood cells.

Humans were assumed to be protected since it had been demonstrated by other scientists that human sera could lyse (digest) most animal retroviruses. A second reason was that in animal models, disease-causing retroviruses, when present, reproduced high levels that were easy to find [4]. It was assumed that the same would be true in humans. Gallo countered these arguments by noting that human sera had only been tested against a few animal retroviruses. So it was an open issue whether they lysed all of them. Moreover, the efficiency of the process might not preclude some cells from being infected. As to the animal models with high levels of virus, Gallo noticed that most of those animal models were selected as lab tools because of their high rate of disease and associated high levels of virus, all irrelevant to the possibility of human retroviruses.

In the midst of all these important developments, Gallo's logic led him to redirect his research thrusts in more fruitful directions and entered the field of Retrovirology, with the ultimate long term goal of unraveling the connection between RNA viruses and cancer induction in humans [1].

The development of new biomolecular assay tools

Gallo first studied animal retroviruses as model systems that might teach him various fundamentals of how cancer occurs, how cancer occurs in humans, even if human cancers were never caused by retrovirus (according to the conventional wisdom of the time). However, soon after beginning his studies with animal retroviruses, Gallo became very suspicious that humans were also likely targets of retroviruses and went after this lonely task with the tenacity to prove himself right against an unconvinced scientific community. So he undertook the venture of seeking, characterizing, and comparing reverse transcriptase enzymes of many different retroviruses, in many different infected animal species as his reference systems. His intermediate goal was to develop sensitive and specific assays for detecting reverse transcriptases in any mammalian system, and for differentiating reverse transcriptases from DNA polymerases which mimic reverse transcriptases. DNA polymerases are enzymes found in cells that catalyse the synthesis of DNA. Reverse transcriptase is a special kind of DNA polymerase carried by all retroviruses.

Then, he used those same assays to search for the presence of reverse transcriptases in human cancer cells. Such a finding would support the claim that at least some retroviruses could be infecting and possibly causing cancer in humans.

Between 1970 and 1972, Gallo's team systematically and painstakingly developed the most sensitive and specific assays ever for detecting all kinds of species-specific reverse transcriptase enzymes, under a well organized plan, and liberally made them available to the scientific community around the globe. These assays were never patented, although at the time, discoveries in molecular biology were already translating into patentable innovations. More importantly, they also advanced the state of the art for human retrovirus detection [3].

In 1972, armed with these assays, Dr. M. Sarngadharan (affectionately called Sarang by everybody in the lab) and Dr. Marvin Reitz, both on Gallo's team, detected the presence of reverse transcriptase in human blood cells from a patient with lymphocytic leukemia [5]. This was an electrifying finding. The footprint of a retrovirus was finally detected in a human cancer sample. Because of this and several other simultaneous observations, suddenly, Gallo's work deservedly got the enthusiastic attention of top administrators at the National Cancer Institute.

The publication of Gallo's finding, suggesting the presence of a reverse transcriptase molecule in human leukemic cells, attracted little attention. By itself, the finding was exciting. But it was insufficient to clinch the case of a cancer-causing human retrovirus. Three important questions were still looking for answers: 1) What was the nature and origin of this reverse transcriptase? 2) What kind of retrovirus could produce

such a particular reverse transcriptase? and, 3) What was the role of the alleged retrovirus in human cancer causation? Finding the answers obviously required isolation and characterization of the retrovirus to which the reverse transcriptase belonged.

However, before one could start isolating and characterizing the first human retrovirus, one had to have significant amounts of live virus on hand. This meant first solving the problem of keeping the retrovirus replicating in cells. It also meant discovering how to grow any retrovirus inside human cells within a cell culture laboratory system, a knowledge not available at the time. To accomplish that task, Gallo had to seek, identify, and use growth factors that could keep leukemic white blood cells growing in a continuous culture, or at least long enough to allow the presumed human retrovirus to replicate in sufficient quantities. This was necessary in order to be able to prove the presence of the retrovirus, to be able to identify its features, and to be able to transmit it to other permanently growing cell lines. Leukemia was still Gallo's primary target disease for his research during this time.

The development of an immortalized leukemic cell line

Gallo assigned the search for a growth factor to scientists Robert Gallagher and Zaki Salahuddin. From the very start, Gallagher suggested that the best chance of finding such a factor would be to work with human embryo tissues, whose normal development appeared to depend both upon the release -and uptake- of growth factors. This suggestion made good sense, so a corresponding approach was implemented, and the search began. One day in 1973, Gallagher and Salahuddin finally met with success when they managed to extract a potent growth factor from a culture fluid in which one of their embryo tissues was growing. It was through regular infusions of this growth factor that they could keep a population of human myeloid leukemic cells (or granulocytic leukemias which occur in bone marrow cells) in continuous growth. At that time, there were some known growth factors for these kinds of cells, but their activity was limited to the growth of these cells in small numbers and/or for short periods of time on a solid surface.

One of these leukemic cell populations proved promising in that it did test positive for reverse transcriptase, signaling the presence of a retrovirus. This particular cell population, named the HL-23 cell line, remained strictly dependent upon regular infusions of the extracted growth factor for continuous growth. Surprisingly, however, another myeloid leukemic cell population, the HL-60 cell line, became spontaneously immortalized, forcing cells to replicate uncontrollably. In other words, it kept on growing and reproducing itself without the need of regular growth factor infusions, only ever requiring periodic additions of nutrient fluid. This second cell line, however, never tested positive for reverse transcriptase. Obviously, the HL-60 cell line was transformed by an unknown mechanism while the HL-23 cell line was reproducing the virus, but without becoming immortalized.

That immortalized HL-60 cell line was immediately made available to other scientist throughout the globe, and to this day remains a tool for many kinds of biochemical and biomolecular studies against this particular leukemic cell species. Indeed, it was the first time that this kind of cell (known as myeloid or granulocytic) was ever grown in the laboratory in a continuous culture.

Disaster strikes

It happened without warning one Monday morning. The freezer, where both the stock of fetal cells producing the growth factor and the stock of the extracted growth factor itself were stored, was left unplugged over an entire weekend [1]. Feelings of dismay, anger, and despair swept the lab. Everything was lost. Gone! Without growth factor, the HL-23 leukemic cell line could not be kept alive. Without growth factor, the virus contained in the HL-23 leukemic cell line could not be kept replicating. Meaning that without that growth factor, the HL-23 leukemic cell line and its virus could not be made available to other scientists for independent verification studies. It was a staggering blow.

Once the initial shock from the loss was over, the search for the same, or a similar growth factor, started all over again. By this time, however, embryonic research had become a hot political issue and fetal specimens were difficult, if not impossible to get anymore. Yet, despite those difficulties, dozens of specimens were obtained and tested in the hope of recovering the badly needed growth factor from a new fetal source. These efforts continued for almost a year, unfortunately to no avail, leaving Gallo to accept the painful reality that the original growth factor was now irretrievably lost and that the prospects of finding a substitute from another fetal source were practically non-existent.

Disaster strikes again

Simultaneous to the ongoing search for a growth factor from a new fetal source, Gallo organized a parallel

search for human or animal cell lines that would continuously grow in culture and could become infected by the virus of the HL-23 leukemic cell line. Almost any cultivable cell line was tried, but the virus stubbornly refused to grow in any of them, evidenced by the discouraging negative reverse transcriptase assays performed time and time again.

Then, two independent pairs in Gallo's lab were given the same goal, hoping that they would bring an end to the problem. One pair was Robin Weiss with Natalie Teich, who came from England as experts for culturing animal viruses. The other pair was Robert Gallagher with Zaki Salahuddin, already experienced in using a variety of animal cell lines. Together, they achieved the unexpected. Their assays tested positive for reverse transcriptase activity, sample after sample, a firm evidence that the retrovirus had transferred from the HL-23 line and had, in fact, infected the animal cell lines. Samples were immediately sent to scientists in other labs for independent examination and confirmation. Electron microscopy confirmed the presence of a retrovirus with the same structure known to cause leukemia in many animal species.

But scientists, who had received and examined the samples sent to them for independent confirmation, reported back that Gallo's findings were nothing but a case of mistaken identity. Their own studies had revealed a contamination of the samples by a cocktail of 3 primate retroviruses, the gibbon ape virus, the woolly monkey virus, and the baboon virus. This composite contamination was most puzzling as Gallo's lab never even possessed those 3 primate viruses to experiment with. In fact, Gallo and his co-workers were themselves already coming to the same conclusions of contamination.

Also about this same time (the mid-1970s), Max Essex, from Harvard University, undertook the study of cat leukemia as an infectious disease transmitted by a virus, which was spread through sexual contact and saliva. The virus was shown to suppress the feline immune system.

The discovery of Interleukin-2

When the search for the recovery of a growth factor from new human fetal sources failed, Gallo turned his attention elsewhere. Phytohemagglutinin (PHA) is a plant extract with the strange ability to agglutinate red blood cells and to stimulate normal white blood cells, specifically lymphocytes, so as to replicate once or twice in culture. Gallo wondered whether PHA-stimulated T-lymphocytes released any growth factors and found that, in fact, they did. He soon realized, however, that one of these factors, known by the name GM-CSF, had already been discovered by other investigators, but whose work did not show they were derived from T-lymphocytes.

Gallo pushed on and in 1974, he and his co-workers, Alan Wu and Joan Prival, pinpointed T-lymphocytes as the main source of GM-CSF. This was one of the main demonstrations that cells of one lineage (lymphocytes) could regulate locally the cells of another lineage (GM-CSF has its effects on promoting maturation of cells of the myeloid lineage). Although the phenomenon of one cell type regulating another was known for hormones, it was not known for locally produced cellular regulators. These locally produced cellular regulators are today generically called cytokines. And if made by lymphocytes, are sometimes called lymphokines.

Doris Morgan, a post-doctoral fellow at Gallo's lab, had a PHA-stimulated blood cell culture growing for long periods against all conventional wisdom, as T-cells were at the time not known to grow in culture past a few cells divisions. It was quickly discovered that T-lymphocytes in culture made, and actually released, several growth factors, one of which would keep the T-lymphocytes growing for long periods. But up until that time there was no known growth factor for T-lymphocytes, and no such factor was suspected to even exist. This, then, was truly a major discovery. At last! A new growth factor had been found from which T-lymphocytes could grow more T-lymphocytes in long-term culture. They reported their findings in Science in 1976 [6]. Basically, what Morgan, Ruscetti, and Gallo had discovered, was a T-cell growth factor, which allows long-term *in vitro* cultivation of human T-cells and ultimately from which human retroviral infection can be detected using a reverse transcriptase assay. That revolutionized the technology for human retrovirus cultivation [4].

By 1977, this new growth factor, which came to be known as Interleukin-2, was more fully characterized in Gallo's lab by Francis Ruscetti. By 1980, the growth factor was purified in Gallo's lab by James Mier. In fact, Interleukin-2 was such an important tool, that it quickly attracted the attention of other scientists.

No one knew it, but another important discovery was also waiting in the wings. A number of leukemic T-lymphocytes were found by Bernard Poiesz in Gallo's lab, to respond directly to Interleukin-2 and grow in long-term culture without prior stimulation from PHA. It would be from these very leukemic T-lymphocytes, stimulated with Interleukin-2 in culture, that Gallo's group would soon at last discover what other scientists scoffed at for so long, the first human retrovirus.

The discovery of the first human retrovirus (the first leukemia virus)

The first cancer-causing RNA viruses were found in chickens around 1910 by Peyton Rous and proved to be infectious. Fortunately, however, cancer-causing retroviruses are less commonly infectious in mammals. It is not surprising, therefore, that when other scientists tried to verify Rous' experiments using mammals, they never succeeded. Based on such negative evidence, clinicians rejected the notion of cancer as a communicable human disease and, in turn, rejected the idea of cancer-causing retroviruses in mammals.

Ludwik Gross was one among a handful of scientists left in the 1950s, who persisted and finally proved that retroviruses are transmissible, albeit rarely, in mice. He accomplished this by inducing leukemia and lymphomas in the laboratory, and showed that retroviruses could be transmitted especially when newborn mice were infected. Following Gross' findings, a whole variety of cancer-causing retroviruses in mammals were later discovered by other investigators.

The next breakthrough came a decade later when William Jarrett showed that transmissibility of cancers by retroviruses was not limited to laboratory animals, but could be observed in feline species under natural conditions. Spurred by all these findings, a Virus Cancer Program was organized in the late 1960s by the National Cancer Institute to hunt for cancer-causing retroviruses in humans. Efforts were renewed and soon they were able to prove the existence of cancer-causing retroviruses in cows and primates. More importantly, they showed that these viruses were capable of intra -and inter- species infection in those animals as well. Despite that and other advances in animal retroviruses, the Virus Cancer Program was unfortunately canceled in the late 1970s, after failing in its goal to substantiate the existence of cancer-causing retroviruses in humans.

Only Gallo stubbornly refused to let go and pressed on, even as others halted this line of investigation entirely. Moreover, by this time there were at least a dozen false starts by investigators all over the world who had earlier thought they had discovered human retroviruses, only to later realize that an experimental flaw had invalidated their work.

With sensitive biomolecular assays to detect any one kind of reverse transcriptase activity and Interleukin-2 to keep the leukemic T-lymphocytes growing in long-term culture (which -if infected with a retrovirus- might continually produce viruses), and the fact that T-lymphocytes were now known to be a major target of retroviruses in a variety of animal models, Gallo set out to prove he was right about the existence of a human retrovirus.

First, leukemic T-lymphocytes were stimulated with Interleukin-2 and grown in culture, expecting to release reverse transcriptases [6]. Reverse transcriptases were then detected by Bernard Poiesz in the fluid of the culture. Specific antibodies both to normal human polymerases (alpha, beta, and gamma) and to different animal reverse transcriptases, were also used by Poiesz. This then proved that the reverse transcriptase detected was neither a normal human cellular enzyme, nor a contaminant from a common laboratory animal retrovirus; rather, it was a novel molecular species [7,8]. This novel reverse transcriptase species was, in turn, purified and shown to possess all the properties of a viral enzyme.

The presence of viral structures in the fluid of the culture was next demonstrated by electron microscopy. The absence of animal retroviruses in the nutrient broth, feeding the cultured cells, was confirmed as well by means of specific molecular assays so as to exclude contamination by animal retroviruses. Viral particles were identified in, and extracted from, the fluid of the culture [4]. The major protein core component of the viral particles was isolated, purified, and tested with various antibodies, and the sequence of its amino acid components recorded, proving that the virus was novel by both criteria [8].

Additionally, the presence of reverse transcriptase was sought, and found, in fresh blood from leukemic patients. It was then shown to be identical to that released by the cultured leukemic T-lymphocytes. Viral genes were also sought by the technique of molecular hybridization, using nucleic acid probes, and found integrated in the genome of T-lymphocytes which were drawn from leukemic patients. This technique allowed direct identification of homologous genetic segments (corresponding in basic type of structure) through molecular stranding. Those results, obtained by Marv Reitz, showed that the virus was not an animal virus contaminant. Specific antibodies against specific viral components (reverse transcriptase and core protein) were then sought and found by Marjorie Robert-Guroff in fresh blood of leukemic patients, an indication of infectivity. Finally, the same new virus was independently isolated from other leukemic patients too. But mere detection and isolation of a new virus means little by itself. Understandably then, when Poiesz first reported the detection of the retrovirus, in his interview for this paper, Gallo recalls his response was that "this is just the beginning of the beginning of the beginning". They needed still to prove that the virus…

- was a novel RNA species,
- was infectious,
- was integrating into the DNA of human cells,
- was present not just in one patient, but to some extent in the human population,
- was the cause of the disease (a particular leukemia),

– could grow in culture from where it could be re-isolated, and even
– could be re-isolated from another sample from the same patient.

Only when all of these tasks were completed, would Gallo allow publication of the discovery. After an all out team effort, after over a year of hard work, and after utilizing the involvement of many of Gallo's investigators, they had discovered and characterized the first human retrovirus ever!

The discovery of this first human retrovirus by Gallo, named HTLV-1 (Human T-cell Leukemia Virus), was accomplished in late 1979 and was presented to peers at scientific meetings in 1979. The first paper was submitted in mid-1980 to the Proceedings of the U.S. Academy of Science and was published in December 1980, under the title "Detection And Isolation Of Type C Retrovirus Particles From The Cultured Lymphocytes Of A Patient With Cutaneous T-Cell Lymphoma". Other papers were also submitted to important specialty journals at about the same time.

Despite the inevitable initial skepticism given over numerous scientific failures in the past, including Gallo's own, all critics were at last convinced that human retroviruses did indeed exist. Soon the existence of HTLV-1 became irrefutable in view of all the overwhelming experimental evidence published.

It is of interest to note, however, that when leukemia caused by the virus finally develops in the patient, usually neither the HTLV virus nor the HTLV proteins can be detected, meaning that the virus, rarely replicates in the actual human subject (*in vivo*). Viral detection is only possible when the T-lymphocytes are properly cultured *in vitro* [6,9]. This is one reason why the detection of a human retrovirus proved a most difficult task indeed.

In 1981, a Japanese group led by the late Yohei Ito reported the isolation of HTLV-1, about a year after the initial Gallo publication on the first human retrovirus, and provided the first independent confirmation of Gallo's discovery. The first independent isolation in the U.S. was achieved by Dani Bolognesi at Duke University. By 1982, no serious scientist would doubt the existence of human retroviruses. It should also be said that years later, those same specific antibody tests developed by Gallo's group to detect the presence of HTLV-1 proteins, would be used in American and Japanese blood banks to screen them against the leukemia virus, protecting transfusion recipients against contaminated blood.

The discovery of the second human retrovirus

In the spring of 1981, Gallo attended a meeting on leukemia in Venice. There cell biologist David Golde of UCLA presented his work on a very unusual, permanently growing T-lymphocyte line, from the spleen tissue of a patient with a rare leukemia called hairy-cell leukemia. This particular line was making lymphokines which Golde had patented and later sold those rights to Genetic Institute. Gallo was quick to realize the significance of Golde's cell line, given that animal -and by now human T-lymphocytes of the type Golde described- generally grow in culture, become immortalized, and make various lymphokines usually when they are transformed by a retrovirus. Armed with his experience on HTLV-1, Gallo suggested at the meeting that another retrovirus could be transforming T-lymphocytes into the hairy-cell leukemia species, allowing them to grow permanently in culture. In fact, the manifest differences between lymphocytic leukemia and hairy-cell leukemia were suggestive that a new retrovirus, not HTLV-1, but most likely a variant, might be causing the latter disease. Gallo further suggested to Golde that it might be most interesting to start looking for another retrovirus at work, so he requested access to the cell line. Because of the patient issues involved, this last suggestion was not greeted with particular enthusiasm and Gallo was refused access and collaboration at that time.

Presumably, Golde then, equipped with Gallo's suggestion, went back to his lab to work on proving that suggestion single-handedly. Six months later, however, Golde changed his mind after unsuccessfully trying to isolate the virus on his own and he asked Gallo for collaboration, offering Gallo the media in which those cells were being grown. Although it is extremely difficult to isolate these human retroviruses from media, Gallo's team was successful in doing just that. They succeeded in isolating and characterizing another new retrovirus [2], which they named HTLV-2. They also showed that the genetic homology between HTLV-2 and HTLV-1 was limited to about 50%. Golde co-authored the publication of the discovery. Kalyanaraman, a young postdoctoral fellow collaborating with Gallo, conducted the immune assays which discriminated HTLV-2 from other retroviruses, and got first authorship for this effort. The discovery of HTLV-2 was soon confirmed independently by others [10].

Contrary to its predecessor (HTLV-1), HTLV-2 infections were discovered to be prevalent among drug addicts in the United States and Europe. Other studies indicated that similar retroviruses were frequent in old world monkeys and apes, and that the origin of the HTLVs in humans was likely the result of a very ancient spread (thousands of years ago) from these primates to mankind.

Gallo later collaborated with Harvard Professor

Max Essex to investigate the role of the HTLV retroviruses in causing immune suppression in humans. The evidence did show that these viruses weaken the immune system of human patients and, almost overnight, Essex's studies on cat leukemia inevitably become mainstream human cancer research. If cats were severely immunosuppressed by animal retroviruses, then why couldn't humans become severely immunosuppressed by human retroviruses? [3]. Takatsuki's prior observations in Japan had already shown a positive indication of human immunosuppression caused by both the leukemia-inducing HTLV-1 virus, and through its effects on T-cells.

Interviewed for this paper, Dr. Essex states "The first time I remember having serious discussions with Bob (Gallo) was in 1971-1972. That was just about the time all the evidence came in showing that cat retroviruses were clearly linked to naturally occurring leukemias (first evidence of this was published by Dr. Bill Jarrett, renowned virologist from Glasgow, Scotland) and that sort of kept alive the idea that such retroviruses might be in people for naturally occurring diseases. About that time we (Essex's group) published the first papers that such viruses could cause immune suppression. And Gallo was really, really excited about that."

Meantime, outside the realm of science, during the second half of the twentieth century, people were erroneously led to believe that infectious diseases were being brought under control and would no longer pose a threat to mankind. Certainly not to the industrialized world. This misplaced faith in the powers of medical science was shattered almost overnight in the early 1980s by the AIDS outbreak in the United States. The outbreak was first detected among young homosexual men in the New York, Los Angeles, and San Francisco areas. Yet, in a sense, there was both a prelude (namely, rising venereal disease infections), and a post-script (the re-emergence of tuberculosis).

Few scientists were willing to take chances and many kept their distance from AIDS with its unconventional epidemic profile, its long latency period, its unforgiving nature (no recoveries), its aggressive spread, and its theoretical danger to those handling patients and samples, choosing instead less urgent and less risky medical projects to work on. Scientists also foresaw the wave of high public despair coming, due to the wild spread of the disease. The extreme public demands and expectations for quick scientific progress put rather high pressure on the entire health care establishment for assertive action [10].

Gallo himself had to make a personal decision too. With a number of important discoveries already to his credit, it would have been safe to do nothing, watch the events unfold, and just give informed advice to all those seeking it. Dr. Phil Markham, interviewed, remembers: "He (Gallo) saw it as a wonderful opportunity." Many believed that a well-deserved Nobel Prize was already in store for the discovery of the first human retrovirus. So at that critical point in his career he had much to lose and very little to gain by entering the uncharted AIDS research arena of that time. Motivated by the challenges in the discoveries that lie ahead, his colleagues were not surprised that he jumped almost immediately into the very heart of AIDS research. On March 18, 1983, Gallo sent a memo to the NCI Director and announced his willingness to get involved in AIDS research at a time when his lab was being inundated with volumes of requests for help, reagents, and advice, stemming from his work in Human Retrovirology [1]. The mail and the phone calls were unending. Still, he wrote in a memo to his NIH superiors that he was tempted by his own competitive spirit to find out what was going on in AIDS.

By getting into AIDS research from almost the very start of the outbreak, Gallo brought much to the table, such as his previous knowledge on human retroviruses which proved critical [1]. Without it, progress on AIDS research would have stayed years behind from where it is now. Dr. Farley Cleghorn who began his career in Gallo's lab as a Research Fellow in Viral Epidemiology (personal communication to this author) believes "When you look at the scientific record, the scientific record clearly shows the body of work that led Bob to the discovery of HIV includes the discovery of HTLV, includes T-cell growth factor (IL-2); without it he would never have found HIV. None of that could have happened. We would still be back in 1985 now if all we had was the discovery of (the French isolate) LAV. The discovery of the first human retrovirus (HTLV-1) was a door that opened, that allowed a truck to get through." In fact, never was so much accomplished so quickly, over a problem this difficult. Especially if one considers the following:

- that from 1960-1981 there was the silent spread of the disease
- that the disease was identified in 1981
- the epidemiology clarified in 1982
- that a suspected agent was isolated in 1983 and verified as its cause in 1984
- that a blood test for its detection was developed in 1984 and made available world-wide by 1985
- that its causal virus was thoroughly characterized by 1985
- that in 1986 there was the globalization of educational programs
- that also in 1986, we saw the first treatment with AZT
- that an inhibitor for delaying the natural cause of the disease was introduced to medical practice in 1987

– that 1995 brought with it the triple drug treatment (or cocktail).

The late Jonathan Mann called the time between 1983-1985, a period of intense discovery, arguably the fastest movement of medical science from the first detection of a new disease - ever!

When interviewed and asked what does Gallo himself say on the coincidence of timing, the AIDS epidemic beginning just when the field of Human Retrovirology was created, thereby opening a new avenue of exploration, he replied "Like a fairytale. It's like a fairytale. It's hard to believe. What I mean is, yeah, it's like an enormous coincidence. The gods play funny tricks." In the simplest terms, AIDS came almost right after the tools for detecting it were discovered. Otherwise, what might our alternate reality be now?

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