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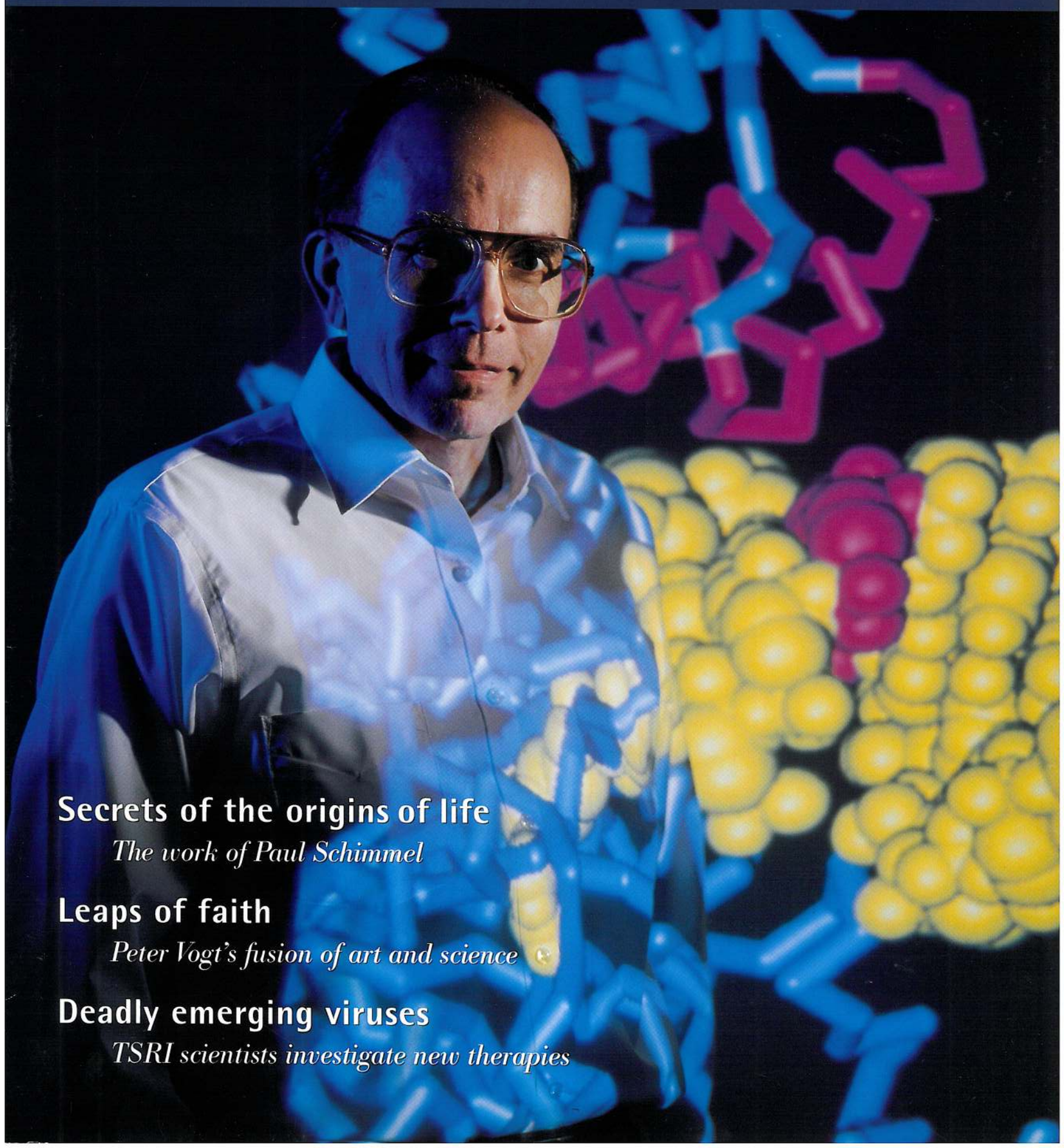

Secrets of the origins of life

The work of Paul Schimmel

Leaps of faith

Peter Vogt's fusion of art and science

Deadly emerging viruses

TSRI scientists investigate new therapies

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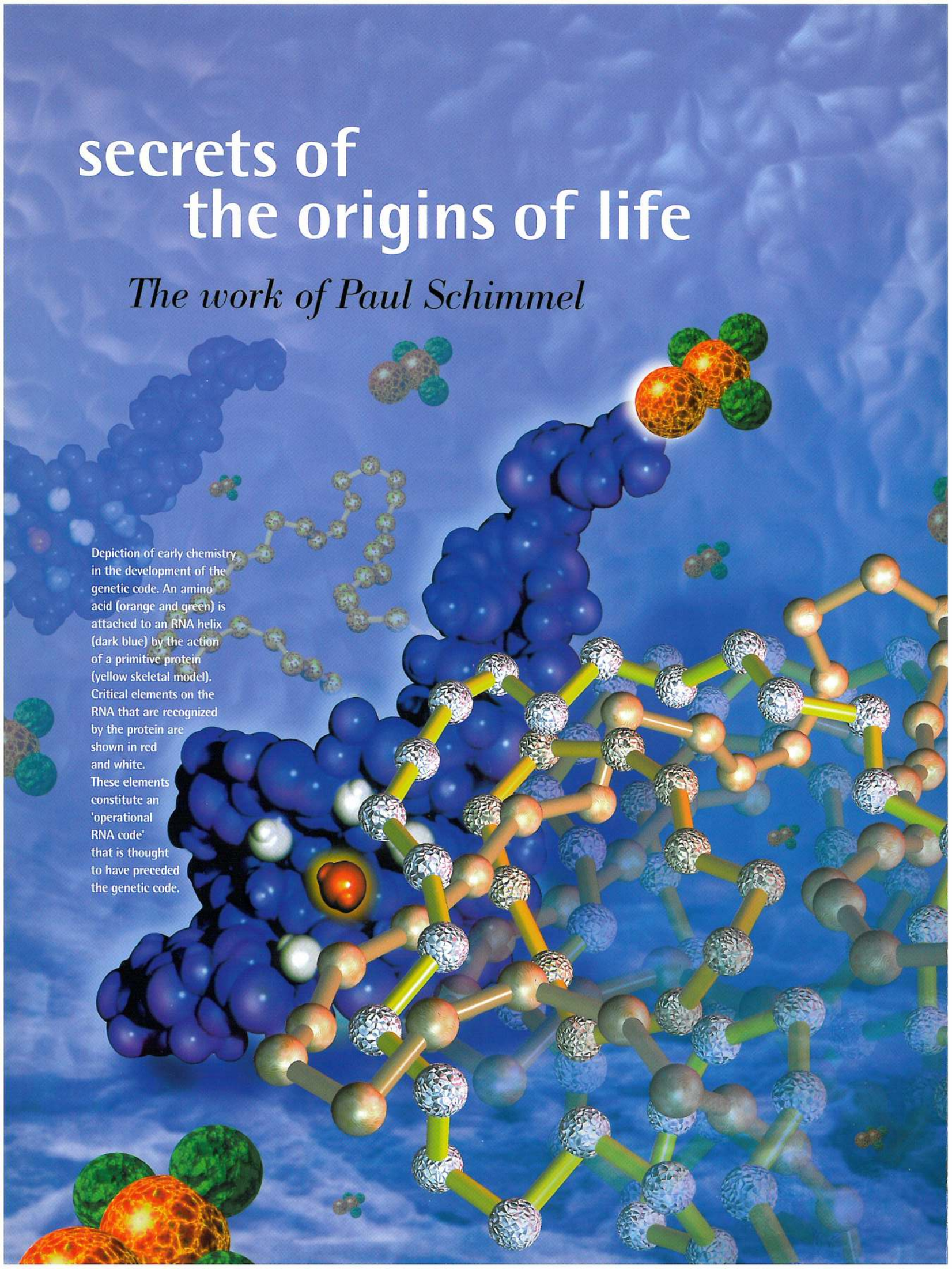


Depiction of early chemistry in the development of the genetic code. An amino acid (orange and green) is attached to an RNA helix (dark blue) by the action of a primitive protein (yellow skeletal model). Critical elements on the RNA that are recognized by the protein are shown in red and white. These elements constitute an 'operational RNA code' that is thought to have preceded the genetic code.

Proteins are the building materials, the pipes and rivets of living cells. Without them, life as we know it would be impossible.

Somewhere deep in primordial time, a tiny creature performed a chemical reaction that changed the world: it somehow read a piece of nucleic acid like RNA or DNA as if it were a computer tape and created a corresponding protein.

Every second, every cell in our bodies reenacts this leap into life, creating robust workhorse proteins out of delicate strands of RNA and DNA. It became known as the "central dogma" of molecular biology: DNA serves as a template for what came to be called "messenger RNA," and this RNA is in turn translated into protein. But ever since this mechanism was proposed in the 1950s, there have been questions. How do cells alive nowadays translate message into protein? How could it first have occurred? And how could knowledge about translation itself be translated into practical drugs and other therapies? These are the questions that have captivated Paul Schimmel, Ph.D., for the entirety of his long and productive research career. Schimmel, who recently arrived to take up a dual appointment as Professor in The Skaggs Institute for Chemical Biology and the Department of Molecular Biology at TSRI, began working on translation more than thirty years ago. His work has placed him "squarely in the middle of the origin of life question," says colleague John Abelson of the California Institute of Technology. It has led to a deeper understanding both of how life works now and how it might have arisen.

"You have to try and figure out how it became that way. That can give you enormous insights into how a cell really works the way it does and how you might intervene."

That Schimmel would become a master of such theoretical questions was unlikely, given his original career path. After attending college at Ohio Wesleyan University, Schimmel moved to Boston to attend medical school. But after less than two years at Tufts, Schimmel moved across the river to Cambridge to attend graduate school at MIT. "I wanted to take graduate level courses in physical chemistry, especially quantum mechanics and statistical mechanics. My father was horrified. His son a med school dropout! But I did it anyway." His relationship with MIT would last for most of his professional life so far — he was a faculty member at MIT for thirty years, most recently the John D. and Catherine T. MacArthur Professor of Biochemistry and Biophysics — before he came to TSRI.

Given Schimmel's interest in the physical end of chemistry, it is unusual that he wound up in a biology department. But in that sense, medical school left its mark. "I wanted to keep my roots in biology. So that's what I did my graduate degree in, even though my advisor was in chemistry." Schimmel went on to work with Chemistry Nobel Laureate Paul Flory at Stanford for a year before returning to MIT.

On The Cover: Paul Schimmel standing with a molecular depiction of components of the operational RNA code for amino acids. The operational RNA code is closely related to the genetic code and its historical development.

STRUCTURE OF BIOLOGICAL MOLECULES

But it is Schimmel's interest in the physical structures of biological molecules that has given his research career its distinct character. For many biologists, the nucleic acid molecules that make up the genetic code are no more than symbols to be analyzed for their content, like words on the page of a book. But for Schimmel, biological molecules — including both nucleic acids and the enzymes that help make proteins — are space-filling physical structures. Schimmel has also distinguished himself by working on both nucleic acids and the proteins produced from their templates. "Most people who work on macromolecules usually choose one or the other," observes

Charles Cantor, who co-authored a 1980 three-volume textbook with Schimmel in biophysical chemistry that has become known as the authoritative book in its field. Scientists who study nucleic acids and those who study proteins "belong to very different cultures," says Cantor. "It requires pretty good breadth to cover them both."

PROTEIN SYNTHESIS

When Schimmel went to work on the translation problem in the middle 1960s, barely anything was known about it. Put simply, protein synthesis looked like this: One group of enzymes "reads" the genetic "words" coded in the DNA of a gene and creates RNA copies of the gene, the so-called "messenger RNA" or "mRNA." Then the message is sent to the cell's protein factory, the ribosome, where another set of enzymes takes the message and translates it into protein, which is made of subunits called amino acids. These ribosomal enzymes read each "letter" of the copied mRNA "word" and seek out the amino acid that corresponds to it in the surrounding intracellular soup. The enzymes find the correct amino acid — there are twenty in all — because without fail it is attached to a molecular label known as a "transfer RNA" or tRNA. The sequence of amino acids in the chain is important: that sequence will determine the structure (and function) of the final protein. So the sequence of letters in the mRNA "words" is critical.

The discovery was hailed by some as a "second genetic code," since presumably it is a different feature on each tRNA that allows it to select the correct amino acid out of the pool of twenty available.

But just as important is how the tRNAs know which amino acid they are supposed to recognize. Protein synthesis would collapse into a shambles if the tRNAs brought the wrong amino acids into the ribosome for assembly. Early in his career, Schimmel set out to determine what it was about the structures of the tRNAs (and the enzymes that manipulate them) that allow them to be so specific. It was not enough, he says, to figure out how the system works now. "You have to try and figure out how it became that way. That can give you enormous insights into how a cell really works the way it does and how you might intervene."

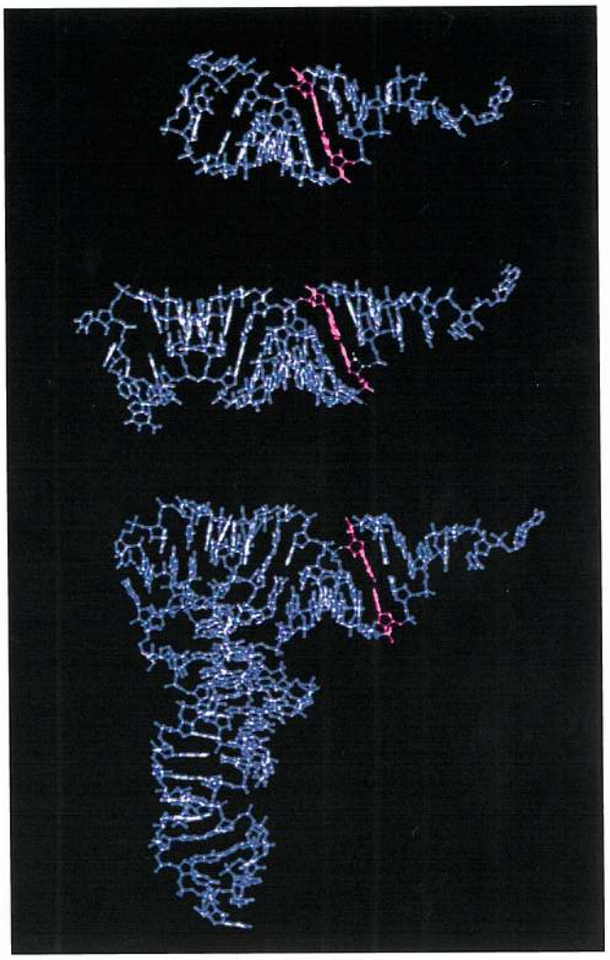
Schimmel applied all the techniques available to him through the early 1970s — this was in the days before cloning and genetic engineering — and got about as far as anyone did. While taking a sabbatical in Santa Barbara, he taught himself molecular biology in its earliest days in 1974 and 1975. His efforts paid off with a series of discoveries. First, in 1981, he cloned and sequenced one of the enzymes that matches up one tRNA with its specific amino acid, alanine. His paper made the cover of *Science.*

A SECOND GENETIC CODE

But he also pursued biochemical and genetic studies of tRNAs, trying to figure out what conferred upon them their exquisite specificity for particular amino acids. Finally, in 1988 there was a major advance: together with postdoctoral fellow Ya-Ming Hou, Schimmel found the feature that distinguished the tRNA for alanine. It was a single pair of RNA subunits (called bases), a "G" and a "U", that are found near the place where alanine is attached. The discovery was hailed by some as a "second genetic code," since presumably it is a different feature on each tRNA that allows it to select the correct amino acid out of the pool of twenty available. But Schimmel prefers to call it an "operational RNA code," since the specificity is created not necessarily by the letters ("GU") — but rather by the overall shape of the tRNA in the vicinity of the crucial pair. "It is the texture of the tRNA that contributes to its matching up with a specific amino acid," says Schimmel.

What was exciting about the operational code is that it was a direct link between protein and RNA, which many scientists believed evolved first, before DNA. Furthermore, Schimmel's later work showed that the entire tRNA molecule was not required to specify a particular protein. This combination, which links proteins to short pieces of RNA, come as close to an understanding of the origin of protein-based life as there is.

"It used to be pejorative even to talk about working on evolution," says Schimmel, "because you are just speculating. But what we have been able to do is design a lot of experiments," which show how things might have actually happened.



Skeletal models of the L-shaped transfer RNA (bottom) and minihelix (middle) and microhelix (top). The two arms of the "L" of the transfer RNA contain, separately, the elements of the genetic code and of the operational RNA code. The minihelix and microhelix come from the top (horizontal) domain of the transfer RNA and they contain elements of the operational RNA code. (The microhelix is a biologically active piece of the minihelix.) The highlighted (magenta) portion is a critical element of the operational RNA code for an amino acid known as alanine.

Schimmel's contributions have multiplied with regard to tRNA and the enzymes that handle it, in particular an enzyme called "tRNA synthetase." He was among the first to establish the modular design of tRNA synthetase by making and studying truncated forms of it. One of the significant results to emerge from his experiments is that one of the enzyme's modules is responsible for selecting the proper tRNA to match with an amino acid, but there is a second, entirely separate module which double-checks the selection and edits it out if it is incorrect.

BIOTECHNOLOGY REVOLUTION

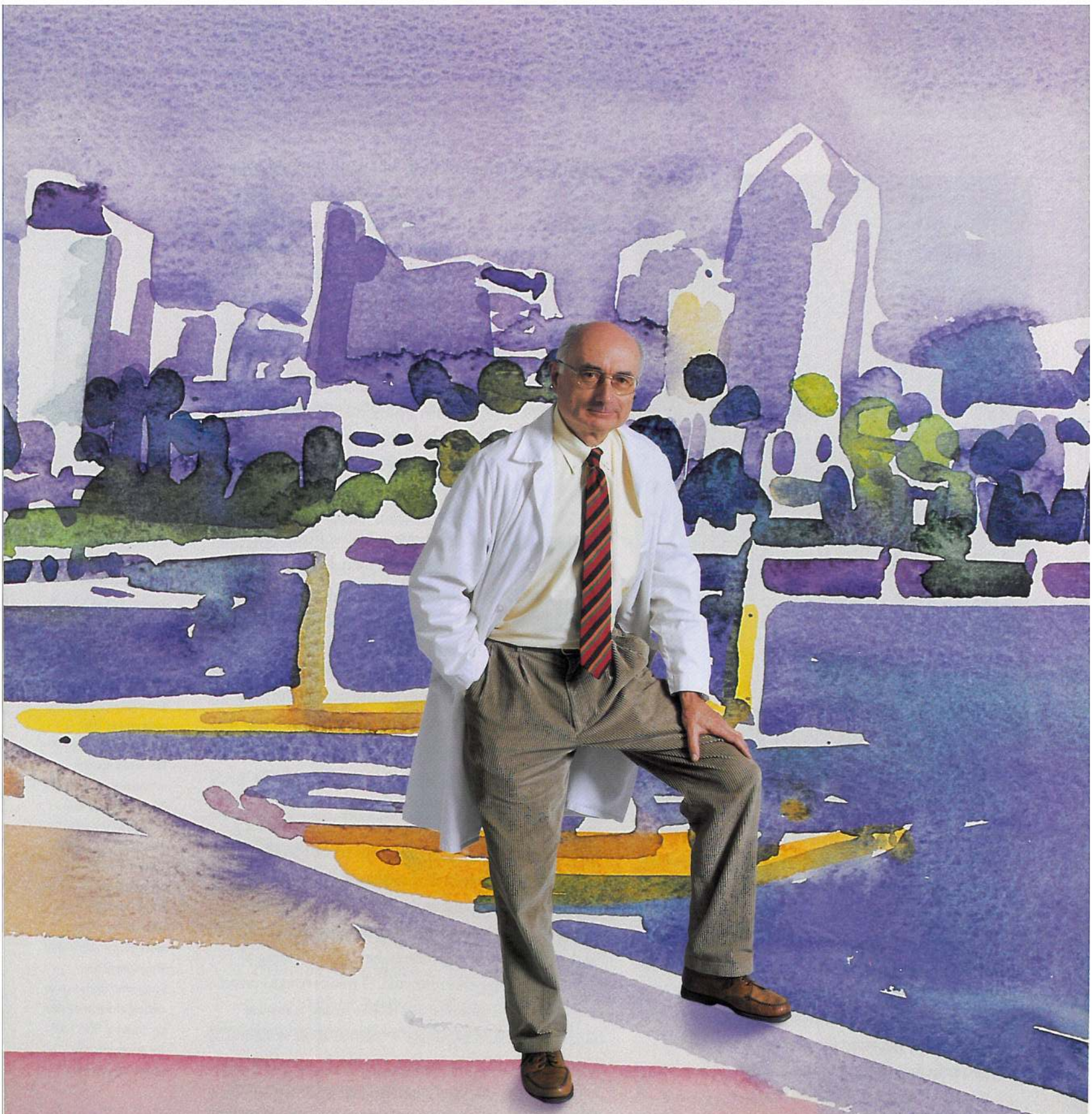
Schimmel's insight that he would have to go to the gene level to understand biology served him in his non-academic pursuits as well. He realized early on what many luminaries of the biological sciences have now come to appreciate: that the scale of research required to make a major impact on therapeutic problems was beyond what the National Institutes of Health were able to fund. "It wasn't so much the commercial side" that attracted Schimmel, he says. "It was more the idea that we were going to transfer technology out of the laboratory. The opportunities were mushrooming."

Schimmel co-founded his first company, Repligen, in 1981 and followed a few years later with Alkermes, a drug delivery company. Neither was closely related in its goals to Schimmel's lab work. Instead, they drew upon his broad scientific vision and from his realization — rare among scientists — that it was wiser to let business goals, rather than scientific curiosity alone, dictate what the scientists in biotech had to do. "[Repligen] was a business from day one, unlike some other biotech companies which offered an excuse to do more science, except with investors' money."

Only with his third company, Cubist Pharmaceuticals, which he co-founded along with then-MIT chemist and current Skaggs Institute director Julius Rebek, Ph.D., did Schimmel get the chance to develop drugs based on what he had learned about tRNA and the enzymes that join tRNA to specific amino acids, enzymes called "tRNA synthetases." These enzymes are different in different species, so they provide an attractive target for drugs that inhibit tRNA synthetase activity in infectious organisms like bacteria but are harmless in their human hosts.

Schimmel's work on tRNA synthetases has led him in a new direction that may be the starting point for a fifth company (he helped one of his former MIT post-doctoral fellows found the fourth one, Amira, in Worcester, Massachusetts, in 1989). "We were able to manipulate synthetase shape and turn on editing activity" in the editing module of the enzyme by providing a prototypical drug-like molecule. "I think we can correct some defects in proteins and RNAs" using a similar method. Traditionally, drugs — such as those designed by Cubist — are inhibitors, meant to block some harmful activity in the cell. But drugs that correct protein and RNA defects would result in a gain, not a loss, of function of the targets. "I have a hunch we can exploit this in a big way," says Schimmel.

The move to TSRI fills Schimmel with a sense of excitement and adventure; it is the intellectual atmosphere that Schimmel finds most attractive. TSRI is a hotbed of research on the origin of life and RNA. And Schimmel is now reunited with his MIT colleague Rebek. For Schimmel, it is like starting a second career at the age of 57.



leaps of faith

Peter Vogt's fusion of art and science

Peter N Vogt

When Peter Vogt, Ph.D., joined The Scripps Research Institute in 1993, his decision confounded nearly everyone he knew.

"First, I made a change relatively late in my career, and second, I left a very secure position at USC as Chairman of the Department of Microbiology. Most people don't do things like that." Vogt, a pioneer in the field of cancer research, laughs easily at the memory, like a man who knows a great secret. "It was a leap of faith."

The secret he knows is this: In a lifetime filled with such leaps of faith, all of which have worked out extraordinarily well, why not go for just one more? He'd been making bold moves ever since the summer of 1950 when as a teenager he woke up one morning and walked away from his East German home to reinvent himself in the West. It was a dangerous step, something he and a friend had been preparing to do for some time. "We knew how to get across the border," Vogt says of that first dramatic journey. "We'd gone across the border in previous summers to travel and look for work. By the time I finished high school, even before that, I knew that I had to leave." He left his entire family behind.

"It wasn't an observational approach ... I liked the precision of it."

Although his parents visited him in West Germany during the 1950s, Vogt never returned to his family home until after the destruction of the Berlin Wall. When they finally came to visit him in California, long after he'd become a prominent oncovirologist and an American citizen, they still did not understand or appreciate how he'd chosen to live his life. Of course, science and California were not Vogt's first choice. He originally told his father he wanted to be a monk. "I was very rebellious as a teenager," he says. "My father wanted me to become a doctor so naturally I told him that I was going to study philosophy and become a monk. But after my first year, I went into science. My father said, 'Well, that's a start.'" His father wasn't the only one caught in the crosswinds of Vogt's dramatically determined choices. The shift to virology happened just as suddenly, after he'd chosen electrophysiology for his doctoral thesis and a zoology professor as his supervisor. It was 1955. Vogt was studying at the University of Würzburg on a scholarship that provided tuition and living expenses wherever he decided to go for graduate studies. That summer he'd come across a book on viruses and it hooked him. The main attraction, aside from the fact that it was a new field and still wide open, was its quantitative approach.

A watercolor skyline of San Diego by Peter Vogt.

EMERGING FIELD OF VIROLOGY

"It wasn't an observational approach like you have in conventional biology. You counted, measured, and used formulas to discover the characteristics of viruses. I liked the precision of it." His supervisor was not as enthralled, especially when Vogt told him he was going to a private institute instead of continuing his graduate studies at the university. It was a risky choice for a young man in Germany to make. There was little active virology research at the time and only one place where it was actually studied — the Max-Planck-Institute of Virology in Tübingen. Moreover, private institutes were not appreciated by German academics, a prejudice that emerged with full force when graduate exams — administered by those same university professors — were given. His former supervisor never forgave him. Then there was virology itself, which was concentrated in the study of animal viruses that could only be grown in chicken eggs. There was no significant cell culture methodology and virtually no understanding of how viruses replicated. Two events

coincided to change that. First was the creation of the plaque assay, a new method of producing and assaying animal viruses in a culture — a quantum jump in laboratory technique. Then in 1953, Watson and Crick demonstrated the molecular structure of DNA and the importance of nucleic acids became clear to everyone.

Vogt's first exposure to that groundbreaking discovery happened by chance. He was being interviewed for admission to graduate school, sitting across the desk

"We had no idea how a virus could turn a normal cell into a cancer cell. It took us nearly ten years just to figure out that that was the right question to answer."

from the director of the Max-Planck-Institute who was holding and turning a peculiar plastic model. It was a model of the double helix. "He asked me if I knew what the model was and I had no idea. He smiled and told me I'd better learn it." Vogt did learn it. Two years after that first encounter, he got to meet James Watson. The future Nobel laureate was a visiting lecturer at the Institute and Vogt was impressed by how young he was — just a few years older than Vogt himself. He was impressed with Watson for other reasons. Above all, Watson was an American, an abstract concept that had become thoroughly imprinted on Vogt's personality. In fact, he had already made up his mind to live and work in the United States, a place he had never been before. "You must understand," he says, "I felt very much at home in America without ever coming here." Part of the attraction was from talking with the American scientists who came to Germany to study and lecture. The rest of it came from books. "I read all of Ernest Hemingway and that taught me about the American character and life," he said.

THE MOVE TO AMERICA

In Germany, Vogt quickly found a supporter in Harry Rubin, a Berkeley virologist with a reputation for quirky independence who told him that if he could find a fellowship, he could come to California. Vogt left for America in 1959 and never really looked back.

"I loved the American West, especially California, because it was so completely different from Europe and so open." After admiring the landscape from afar, he now found himself overwhelmed by the actual experience. In response, he began to paint it, immersing himself in its vast canvas of distance and color. Since then, his landscape painting has become a second career that comes close to matching the success of his first. It has certainly captured his soul.

Each autumn, Vogt makes the journey north to Washington State, to a remote island where he closes up his small cottage for the winter after a summer of intermittent painting. It is where the other half of his life — the part he calls his anchor — takes place. It's where he paints some of his landscapes of the American West. Like his love of science, Vogt's painting has its early roots in Germany. At Würzburg, he took Saturday classes with a painter who later became a life-long friend. Vogt apprenticed in his studio, idolized Cezanne as the father of modern painting, and still sees himself as a throwback to an earlier age when painters worked with easel and brush in *plein-air* to capture the light and color of what they saw.

A RELATIONSHIP WITH THE LAND

"It's very old fashioned," he says, "nobody goes out and paints landscapes anymore." Perhaps. But there is more at work here than simple artistic desire, something that Vogt fully admits borders on the mystical and the religious. It is his complete immersion in the land itself, a solitary experience that is quite different from the day-to-day collaborations that are part of modern science. "I love to interact with people but being alone, communicating with the landscape, gives my life a balance in so many ways. I love the California desert landscape to an extent that it helps define who I am. It is a landscape that I understand, that I feel a part of. My relationship with it is very intense." When he describes that relationship, his words are similar to his description of America and the desire of a young man to make the leap to the one place where he knew on faith he belonged. That sense of elation, even gratitude, is reflected in his watercolors with their feeling for shifting shapes and colors of the American West.

His current work at TSRI centers on the expression of

genes in cancer, delving into the fundamental mechanisms of how proteins, acting as the master switches that turn genes off and on, become corrupted and produce cancer cells. Vogt describes them as the last component in a complex sequence of signals that are responsible for cancer behavior. It is the end result of a lifetime spent looking at things no one else was interested in or understood. By the time Vogt made his way to California in the late 1950s, the study of virology had expanded, based partially on the discovery that a tumor virus could transform a normal cell into a cancerous one in culture. Tumor virology was a new field, albeit one that attracted few followers, among them Vogt and his mentor, Harry Rubin. "We worked on avian tumor viruses, they were widely thought of as esoteric with little or no relevance to humans," he says. "Everyone said they were interesting as models but had no direct impact on people. Harry and I thought they provided a really nice niche where we could work in peace."

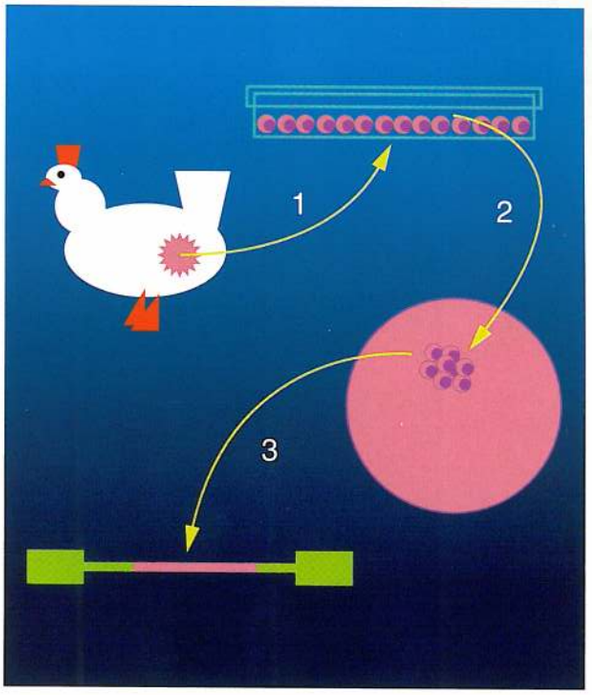
"It's the ideal environment for my work, and I think it points the way to the future of cancer research."

In truth, they had no idea what was important in tumor viruses and what wasn't. To a great extent, they were following Rubin's determined path away from the mainstream, leaping into terra incognita.

FROM A NORMAL CELL TO A CANCEROUS ONE

"We had no idea how a virus could turn a normal cell into a cancer cell. It took us nearly ten years just to figure out the right questions to ask," according to Vogt. He continues, "Initially, few people thought retroviruses were important. But in the late 1960s, Howard Temin and David Baltimore discovered a unique enzyme that turned RNA into DNA and the riddle of retroviral replication was solved. That set us free to look for oncogenicity — the way tumor cells were created from normal cells."

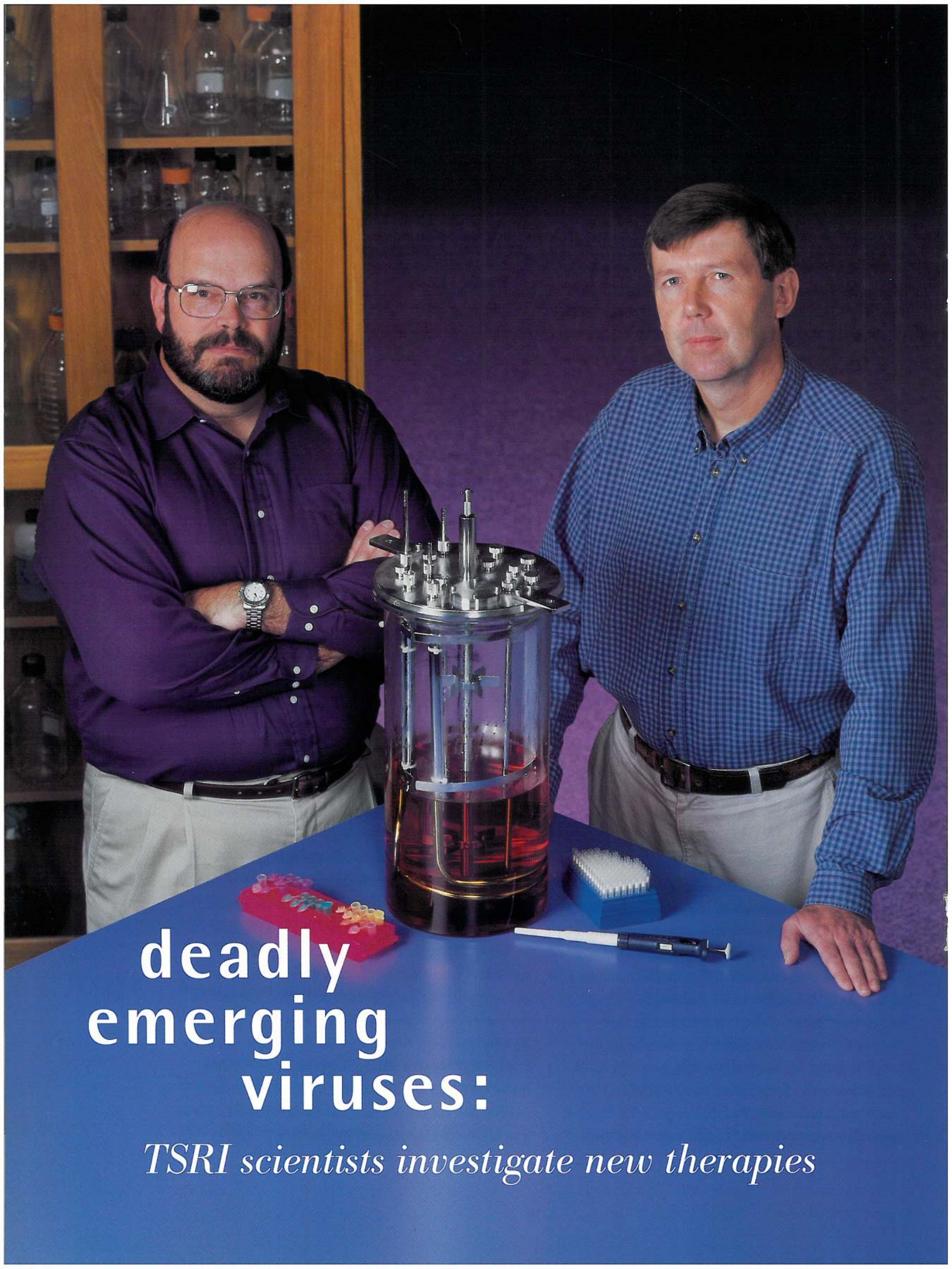
"We found the first tumor-inducing genes in the genetic material of retroviruses. These oncogenes turned out to be hitchhikers that had their origin in the host cell itself. They were once useful and important regulators of



Many cancer genes were first discovered in spontaneous virus-induced tumors in chickens. (1) Tumor cells grown in a plastic dish produce a tumor-causing virus; (2) The viral genetic material, a single molecule of nucleic acid, contains an altered cellular gene that is responsible for the cancer; (3) Such cancer genes found in chicken viruses are very similar in structure and function to oncogenes of human tumors, and much of what we know today about genetic changes in human cancer has its origin in the study of chicken tumor viruses.

cell growth but being incorporated into a virus changed them into dangerous cancer-inducing genes," Vogt explains. He discovered a number of these cancer genes; he still scrutinizes the genetic material of viruses and of cells for new ones. But his main interest now is to discover the mechanisms by which these biological switches so profoundly alter the growth behavior of cells. He also wants to apply the genetic knowledge of cancer to the development of novel therapies that will be effective in the treatment of the disease. Vogt believes that the unique resources of TSRI make this a realistic goal. "Scripps opened my eyes to new worlds," he says. "The fact that we have chemistry and structural biology departments is critical. It was a genius decision to build these into the scientific fabric of the Institute. It's the ideal environment for my work, and I think it points the way to the future of cancer research."

When Vogt made the leap to TSRI in 1993, the Institute had a reputation as a "tough place" where researchers were expected to create their own agendas and to develop their own sources of funding, a discipline that seems custom-made for Vogt, a man who's followed his own lead nearly every day of his life. "People told me I was crazy," he says, still laughing at the reaction. "But I trusted the people at Scripps to help me make the transition, and that trust has been rewarded several times over. I knew it would be the ideal place for me, and it is."



deadly emerging viruses:

TSRI scientists investigate new therapies

Their very names sound sinister: Marburg virus, Hanta virus, Lassa virus, and the famous - if rare - Ebola virus, which was turned into the international superstar of viruses by the best-selling book, "The Hot Zone."

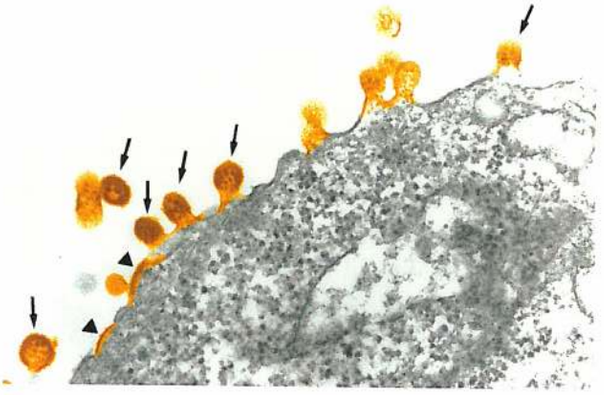
Michael Buchmeier, Ph.D., knows them all. Over the past twenty years, Buchmeier — Associate Professor in the Department of Neuropharmacology at TSRI — has worked on these and other deadly and mysterious viruses. "What we have attempted to do," says Buchmeier, "is to investigate the structure and function of these viruses and use that information to begin to interpret their interactions with the host."

Buchmeier warns that though less well known than Ebola, its companion viruses also present a threat to human beings. "These are not obscure diseases," he says. For instance, Lassa is a hemorrhagic fever virus that initially emerged in Africa in 1967. Lassa fever is similar to Ebola but less severe. It has been estimated to infect up to 200,000 people a year in West Africa. Other emerging viruses such as Hanta virus, which caused a deadly outbreak in the American Southwest in 1993, and which the team is also studying, are endemic throughout three-quarters of the area of the United States, and related viruses are endemic throughout much of Europe and Asia.

BATTLING DEADLY VIRUSES IN THE LAB

Together with Dennis Burton, Ph.D., Professor in the Departments of Immunology and Molecular Biology, Buchmeier is working on laboratory approaches to combating these deadly viruses. Buchmeier, Burton and several other TSRI colleagues received a grant from the National Institutes of Health in 1996 to research immune therapies against Lassa and Ebola. "As it is now, Ebola is not going to wipe out civilization," says Burton. But he adds, "That's not to say some day it won't undergo a few changes or become airborne or combine with some other virus and find some other way of spreading itself."

One of the most frightening qualities of Lassa and Ebola viruses is that the human immune response to them is barely adequate. In a standard viral infection, the body rallies after a few hours or a few days to produce antibodies and specialized killer cells, both of which home in on the attacker and eliminate it. But when Ebola strikes, a human victim typically responds with an immune response so weak as to allow the virus the upper hand in killing the person. Scientists believe that a strong immune response may be a part of what allows those lucky survivors of Ebola to fight off the infection.



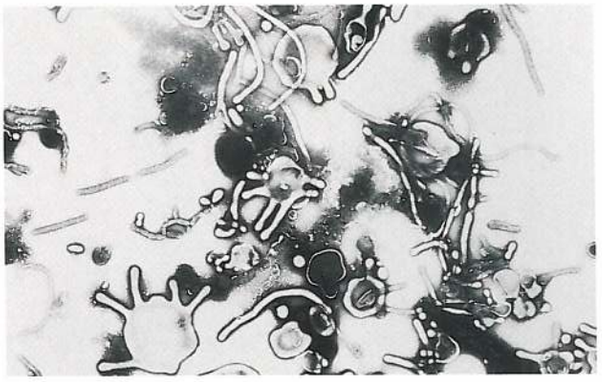
Virus particles budding from infected kidney cells. Arenaviruses including Lassa and LCM virus share a characteristic morphology (arrows) with a dense lipid envelope encircling an interior containing electron dense 20 nm bodies which were originally thought to resemble grains of sand. Budding of particles begins at a patch on the cell membrane (large arrowheads), extrusion of the particle and eventual pinching off to release mature virions.

LOOKING FOR A CURE

Burton is bringing the power of modern science to bear on the side of the victim. Looking in the blood of Ebola survivors, he has found three antibodies that react strongly with Ebola fragments in the test tube. And that gives him a head start on what may someday be a cure for this as yet incurable disease.

Just having the antibodies themselves or serum from recovered victims' blood is not enough, says Burton, since "there aren't many Ebola victims around. There won't be much serum available." Instead, he is using a technique to generate large amounts of Ebola-killing antibodies using genetic engineering.

Left to right: Michael Buchmeier, Ph.D., and Dennis R. Burton, Ph.D.



Concentrated Ebola virus particles exhibit a wide variety of shapes and lengths characteristic of this pathogen.

The first step is finding the antibodies that work against Ebola. From the blood of a handful of Ebola victims, Burton extracted not the antibodies themselves but their genetic blueprints, which he then inserted into bacteria. "We recapitulate the antibody response of the individual in bacteria; then we go in and recover the particular antibodies we're interested in," he explains. The Centers for Disease Control has already confirmed the effectiveness in the test tube of the three antibodies Burton has isolated. He is submitting his work for publication.

The antibodies also point the way for vaccine development, says Burton. "If you know these antibodies will protect [an infected person against Ebola], you know what you're trying to elicit with your vaccine." He expects to begin animal studies soon and to have results within a year.

If Burton's strategy works, it will be a stunning advance —the first protective strategy against Ebola. It will be a welcome novelty indeed for those unfortunate enough to come in contact with the dread disease.

The antibody molecule, one of the most potent anti-viral agents of the human immune system.

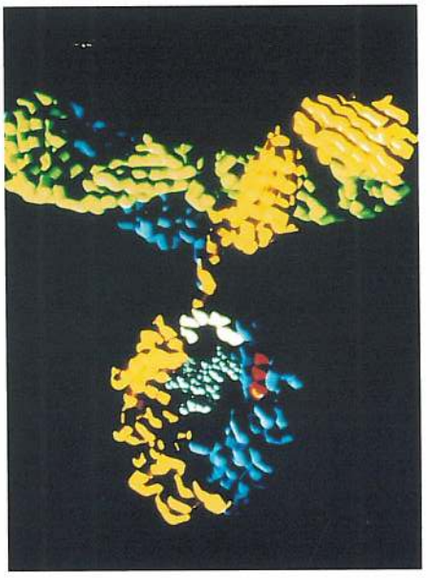
IMMUNE RESPONSE TO DATA

Buchmeier's work parallels that of Burton in a different part of the immune system. Buchmeier works on human cellular immune responses to Lassa. Lassa has infected millions of Africans and a smaller number of western scientists and aid workers; it has killed an unfortunate few.

"We know very little about how people survive Lassa," explains Buchmeier. The antibody response to Lassa seems to come altogether too late to help the victims. The primary way that humans defend themselves against Lassa is for their immune system's "killer cells," the cytotoxic T-lymphocytes or CTLs, to jump into action. If researchers could find a way to mobilize more killer cells in patients already infected with Lassa, they could presumably prevent many deaths. Such a therapy would resemble a vaccine in that it would stimulate the patient's own immune response. But since it would be administered after infection, not before, it would be classified as an immunotherapy. Similar approaches are under development in both HIV and hepatitis-B virus. But in neither case have they reached wide clinical use.

Buchmeier chops up the proteins found in Lassa virus into protein fragments just nine or ten amino acids long. Since it is these fragments that the killer cells recognize, it is essential for scientists to present the right ones to boost a patient's immune response. It takes more than one, but probably fewer than a dozen, says Buchmeier, to prime the patient's immune system to respond vigorously to a Lassa infection. The trick is to find which few, and this is a problem on which Buchmeier and his colleagues at TSRI and at Cytel Corporation have already made great progress.

The next step, once they have identified the key set of fragments, is to obtain blood samples from individuals who have survived Lassa infection and see if the killer cells they contain can be stimulated to attack Lassa virus. If that works, the immune therapy based on Buchmeier's work will enter first animal and then human trials.

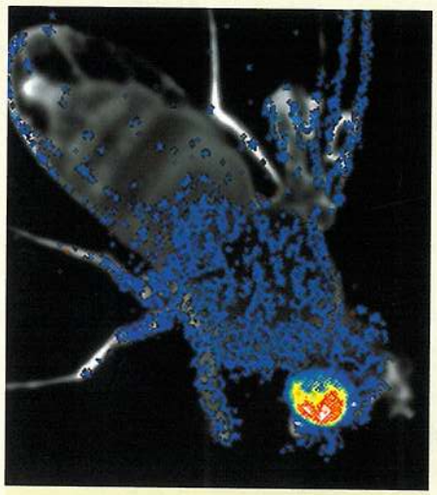


When he thinks about the deadly viruses he works on, one thing gives Mike Buchmeier hope: Lassa and Ebola did not evolve to harm humans. "A deadly human infection is as disastrous to the virus as it is to its human host," he says. It reminds Buchmeier a little bit of the Hippocratic Oath: "First, do no harm." Evolution has somehow led these viruses down a destructive path. Buchmeier and Burton are helping human beings fight back.

Biological Clocks No Longer Found Only in the Brain

A recent discovery by a team of scientists at TSRI and Brandeis University challenges the strongly-held belief that 24-hour rhythms, or biological clocks, are centrally controlled from the brain. Using the fruit fly as a genetic model system to study circadian rhythms, the researchers, led by Steve Kay, Ph.D., Professor, Department of Cell Biology, TSRI, sought to determine if individual body parts would respond to changes in the light/dark cycle without any help from the head. As reported in a recent issue of *Science*, in each separately cultured segment, so-called clock genes turned on and off in unison, according to rhythms set by environmental light manipulations.

These findings demonstrate that time-keeping genes may be running in tissues all over the body and controlled locally in the flies and therefore possibly in mammals, as well. The scientists hope that understanding the location of such clocks in tissues and cells, as well as identifying which genes and proteins make up the biological process, will yield insights into human circadian rhythms that could lead to new strategies for the treatment of disorders associated with jet lag, shift work and seasonal depression.



Bioluminescence in a whole living Drosophila

Per-driven bioluminescence from a per-luc *transgenic* Drosophila *was captured, pseudocolored, and superimposed on a retouched light micrograph of the sample, imaged separately.*

Bioluminescence illuminates several tissues throughout the fly including the eyes, wings, and legs; all of these tissues exhibit rhythmic bioluminescence in culture.

To measure one of the genes that control clocks switching on and off in animals, called *per* for "period," the researchers borrowed some tricks from the world of bioluminescent organisms. They fused the fruit flies' *per* gene DNA to "glow" genes either from jellyfish or fireflies, to make glow-in-the-dark fruit flies. The researchers engineered a transgenic strain of flies that expressed luciferase – the enzyme in fireflies that glows a greenish color in the presence of luciferin – whenever the *per* gene was expressed. To reveal where the gene was expressed, fruit flies were altered to express the gene from jellyfish that produces Green Fluorescent Protein as a marker of *per* sites.

Kay commented, "We found that all these tissues we cultured from the whole animal were glowing on and off, demonstrating that lots of clocks are running throughout the fly, independently of the brain."

Under normal light/dark conditions, the clock genes rhythmically luminesced in each of the cultured segments – head, thorax and abdomen. The tagged *per* genes were especially conspicuous in chemo-sensory cells at the base of hairs on the legs and wings and on the antennae and proboscis. These clocks also ebbed and flowed autonomously in response to light, suggesting that circadian rhythms likely regulate a fruit fly's sense of smell, much as they influence light and pain sensitivity in mammals.

While the authors raise the possibility that this evidence of multiple oscillators challenges the current notions about the role of the brain as the seat of a "master oscillator" that coordinates rhythms throughout the organism, they acknowledge that the brain still retains a certain distinction, even in a fruit fly. In the prolonged absence of light, the head was the only organ in which the *per* genes remained in sync.

A mammalian variant of the Drosophila's *per* gene recently was identified, along with another rhythm-related gene *(Clock)*, both found throughout the body of mice; both *per* and *Clock* make their products in many different mouse tissues. According to Kay, the discovery of many non-brain clocks in fruit flies could well be true for humans. He suggests that, "In this case it might mean that our skin, liver or other peripheral tissues have their own clocks to control these local functions."

Also participating in the research were Jeffrey D. Plautz, Department of Cell Biology, TSRI, University of Virginia, and NSF Center for Biological Timing; and Maki Kaneko of the Department of Biology and NSF Center for Biological Timing, Brandeis University.

The study was funded by the National Institute of Mental Health of the National Institutes of Health, and the National Science Foundation.

Scientists Solve Structure of Active Site of Enzyme that Produces Nitric Oxide

Discovery Suggests Possible New Ways to Design Novel Drugs for Several Human Diseases

Scientists at The Skaggs Institute for Chemical Biology and the Department of Molecular Biology, led by Drs. John Tainer and Elizabeth Getzoff, in collaboration with a team led by Dr. Dennis Stuehr at the Cleveland Clinic, have solved the structure of the active site of the enzyme that regulates the activity of nitric oxide, or NO. Since NO is an unconventional biological signal whose activities range from blood pressure regulation to antimicrobial defense to nervous system information and memory, understanding the structure of the enzyme that produces it is crucial to designing drugs to turn NO on and off. Scientists predict that NO inhibitors may be used to treat such diseases as high blood pressure, septic shock, stroke, cancer, and impotence. Given its role in neurotransmission, NO may have an effect on treating memory disorders and learning.

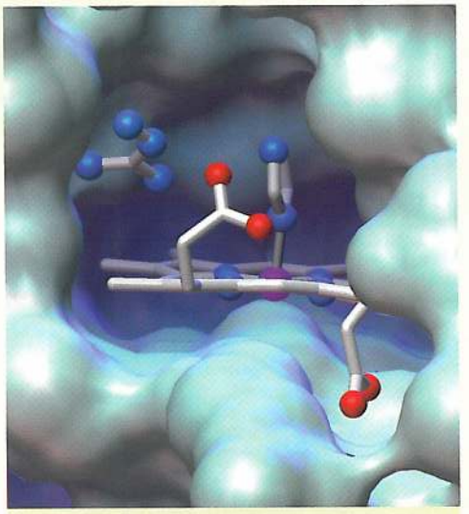
According to Tainer, "Having this structure is the difference between working blind and seeing what you're doing in terms of understanding and drug design."

The structure of this key portion of nitric oxide synthase (NOS) helps researchers understand not only how NO is produced in the body but also how its production is controlled. Nitric oxide is a small, short-lived, inorganic molecule that functions in mammals as an essential chemical messenger for many physiological processes and as a protective poison against pathogens and cancer. At low concentrations it acts as a signal to control blood pressure, prevent blood clotting, transmit nerve impulses in contractile and sensory tissues, process sensory input, form memories, and allow learning.

In contrast, the immune system produces high concentrations of NO and exploits its reactive properties to combat bacteria, intracellular parasites, viruses and tumor cells. Due to its unstable and membrane diffusible nature, NO differs from other neurotransmitters and hormones in that it is not regulated by storage, release or targeted degradation, but rather solely by synthesis.

Because NO acts as a signal in low amounts and a toxin in high amounts, its production is carefully balanced in healthy humans depending on the state of the organism. Pathologies thought to involve too little NO production include hypertension, impotence, arteriosclerosis, and a susceptibility to infection. Diseases linked to excessive NO production include immune-type diabetes, neurotoxicity associated with aneurysm, stroke and reperfusion injury, inflammatory bowel disease, rheumatoid arthritis, cancer, septic shock, multiple sclerosis and transplant rejection.

According to Dr. Solomon Snyder, a neuroscientist at Johns Hopkins University whose research group was the first to clone and sequence NOS, "NO appears to be one of the most important messenger molecules in the body. Excess production appears to cause brain damage from stroke and also inflammatory conditions. Drugs that block the enzyme could be important therapeutically; this breakthrough may allow scientists to begin to design drugs to inhibit it."



The active site heme pocket of iNOS showing the juxtaposed imidazole and amino guanidine inhibitor binding sites.

The chemistry NOS uses to produce NO is complicated and unique in biology, and its structure is completely different from other oxygenase enzymes involved in hormone synthesis and the detoxification of harmful compounds. However, a comparison provides insight into the aspects of these enzymes that are key for the similarities and differences in the reactions they catalyze. According to Tainer, this should aid researchers in reproducing these biological reactions in the laboratory for the design of drugs or other desirable compounds.

Funding for the study was obtained from the Skaggs Institute for Research and the National Institutes of Health.

Promising New Anticancer Drugs from Rare Corals Synthesized in the Laboratory

TSRI scientists have performed the first total chemical synthesis of a number of promising new anticancer compounds, first isolated from rare species of corals and related marine organisms. The team, headed by K.C. Nicolaou, Ph.D., succeeded in assembling these compounds in the laboratory by designing a multistep strategy using simple chemical building blocks such as carvone, an oil readily available from caraway or dill seeds, frequently used as a commodity chemical in perfumes and foods. The scientists hope to produce synthetic analogs for biological screening purposes that could ultimately lead to more effective and safer therapeutic agents than the original compounds.

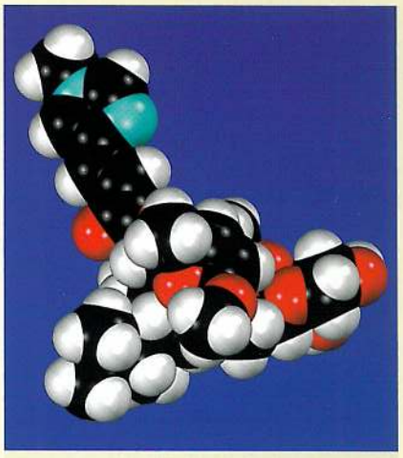
According to Nicolaou, Chairman of the Department of Chemistry, Skaggs Professor of Chemical Biology, and Darlene Shiley Professor at TSRI, and Professor of Chemistry at UCSD, "There is always risk of failure associated with any drug discovery and development program. What we can say for certain at these early stages of the research is that the new substances look very promising in killing cancer cells and now we know how to make and fine-tune them in the laboratory for further biological investigations." The drug discovery and development process often takes more than 10 years and the cost for development frequently reaches several hundred million dollars.

Eleutherobin, one of the novel compounds synthesized, appears similar to the anticancer drug, Taxol,™ in its mechanism of preventing cells from dividing.

Eleutherobin, one of the novel compounds synthesized, appears similar to the anticancer drug, Taxol™ in its mechanism of preventing cells from dividing. It originally was found by W. Fenical and his co-workers at the Scripps Institution of Oceanography, in soft corrals collected from a region of the Indian Ocean near Australia, known as Bennett's Shoal. The compound has shown powerful properties against cancer cells.

Fenical commented, "The stuff was so extraordinarily potent that it was dangerous to handle. You could dilute it a million-fold, and it still killed cells very powerfully."

The second group of substances, sarcodictyins, were first discovered from a Mediterranean stoloniferan coral (sarcodictyon roseum) species in 1987 by an Italian research group led by F. Pietra, but its anticancer activity was only recently reported. The natural scarcity of these compounds coupled with their promising anticancer properties prompted a search for their laboratory production. These chemical syntheses address the issue of supply and open the way for further pharmacological investigations, which may lead to the development of these new substances as chemotherapeutic agents against cancer.



Computer-generated space filling molecular model of eleutherobin.

Scientists have determined that eleutherobin's unusual method of blocking cell division is similar to that of Taxol. Nearly all cells have a complex structure within them called the cytoskeleton, an intricate scaffolding of minute fibers called microtubules. The scaffolding changes according to the functional state of the cell, appearing and disappearing as the microtubules break down and then reassemble. While a number of compounds, including some anticancer agents, can inhibit this reassembly thereby preventing cell division, eleutherobin and Taxol have the opposite effect. Rather than breaking down the internal structure of cells, these compounds paralyze them, making them so stable as to prevent movement, replication or cell division.

The total synthesis of Taxol was achieved by the Nicolaou group in 1994. It has been called a breakthrough treatment for breast and ovarian cancers.

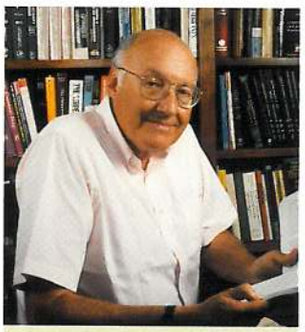
The team that successfully synthesized eleutherobin included postdoctoral fellows Drs. Jinyou Xu, Floris van Delft, Takashi Ohshima, Sanghee Kim, Seijiro Hosokawa, and Dionisios Vourloumis, research associate Tianhu Li, and graduate student Jeff Pfefferkorn. The work was supported by The Skaggs Institute for Chemical Biology, National Institutes of Health, Novartis, and CaPCURE.

Dr. Michael B. A. Oldstone Receives 1997 J. Allyn Taylor International Prize in Medicine

Michael B. A. Oldstone, M.D., has been named a recipient of the 1997 J. Allyn Taylor International Prize in Medicine. Head of TSRI'S Division of Virology and a member of the Department of Neuropharmacology, he has been on staff here since 1966.

The Taylor Prize was awarded to three scientists who have made outstanding contributions to the understanding of virus-host interactions. Others include Dr. Bernard Roizman, University of Chicago; and Dr. Bernard Moss, National Institutes of Health. It is awarded by the John P. Robarts Research Institute, the largest privately-directed medical research facility in Canada.

Oldstone has been recognized for his work in defining how viruses persist by escaping the immune system and characterizing the diseases persistent viral infections cause. He also is being noted for his observations that viruses can cause autoimmune disease. A graduate of the University of Maryland School of Medicine, he began his career at TSRI as a postdoctoral fellow in the Department of Experimental Pathology in 1966. He became an associate member of the Department of Immunopathology in 1972, and a member of the Department of Immunology in 1978. In 1989, he was named head of the Division of Virology, Department of Neuropharmacology, as well as a member of the department.



Michael B. A. Oldstone, recipient of the 1997 J. Allyn Taylor International Prize in Medicine.

Oldstone is the recipient of numerous honors and awards and has served on a variety of prestigious advisory boards. Recently, he was elected to the Institute of Medicine of the National Academy of Sciences.

$5 Million Gift to Fund New Center for Molecular Structure and Design at The Scripps Research Institute

The estate of the late Buddy Taub, a Carlsbad businessman, long-time patient and friend of Scripps, has made a $5 million contribution to TSRI to construct a basic research facility that will house the world's most powerful nuclear magnetic resonance (NMR) instrumentation. The 12,000-square-foot Buddy Taub Center for Molecular Structure and Design will be constructed at the Institute's Lita Annenberg Hazen Science Center on its campus in La Jolla.

NMR is a powerful method for determining three-dimensional images of medically important biological molecules, allowing their structures and shapes to be studied in their natural states, in solution. It also provides information on how those structures change upon interaction with drugs and other key substances, leading scientists to a better understanding of biological function and providing the foundation for computer-based design of novel drugs and vaccines. The NMR facilities at TSRI are among the most sophisticated in the world. With the construction of the new Taub Center and the installation of two additional 600 MHZ spectrometers and an 800 MHZ instrument next year, the TSRI laboratory will be the world's most extensive and best-equipped biomolecular facility. TSRI will purchase a 900 MHZ instrument currently under development which, when installed in the Taub Center, will contain the most advanced magnet in existence.

The specially designed building, at an anticipated cost of $13 million, is expected to be completed by the end of 1998.

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