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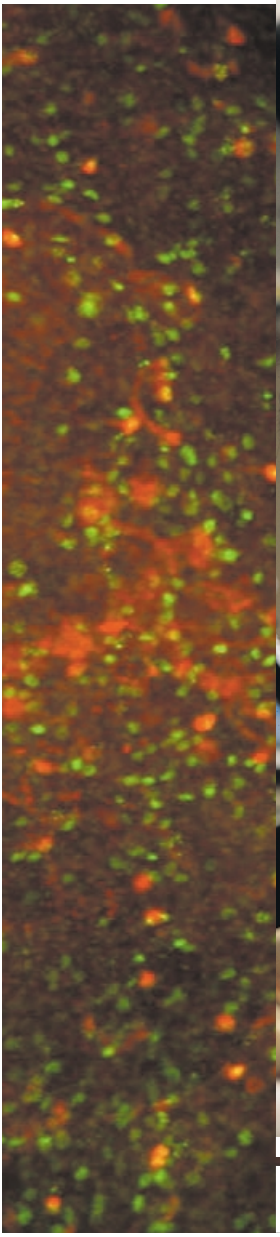

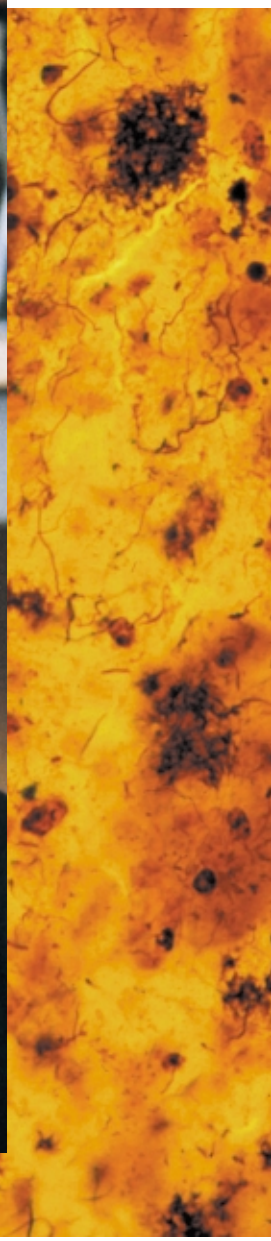

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On the Cover

With the help of an automated microinjector, Alejandro Sánchez Alvarado gently guides a glass needle filled with DNA-labeling molecules into an anesthetized flatworm under a microscope.

Cover photo of Alejandro Sánchez Alvarado: *William K. Geiger*

By studying how cells stick to blood vessels and other surfaces, Laura Kiessling is trying to understand inflammation and Alzheimer's disease.

Cover photo of Laura Kiessling: *Jeff Miller*

Editor's Note

How do cells stick together? How do they combine just right to make a living thing? Fundamental biological mysteries such as these keep thousands of scientists busy around the clock. As humans, we are all concerned with how living things—like our bodies—keep themselves running smoothly, and scientists yearn to know how to fix problems that worsen our health.

The research you'll read about in this issue of *Findings* is fueled by the National Institute of General Medical Sciences, a component of the National Institutes of Health. The stories are examples of the cutting-edge science that is paid for by U.S. tax dollars. *Findings* wants to show you the excitement scientists feel every day in their labs, where they search for answers to endless riddles about the human body. And they have fun doing it! Check out the NIGMS Web site for more information—and for the solution to the crossword puzzle on the last page!

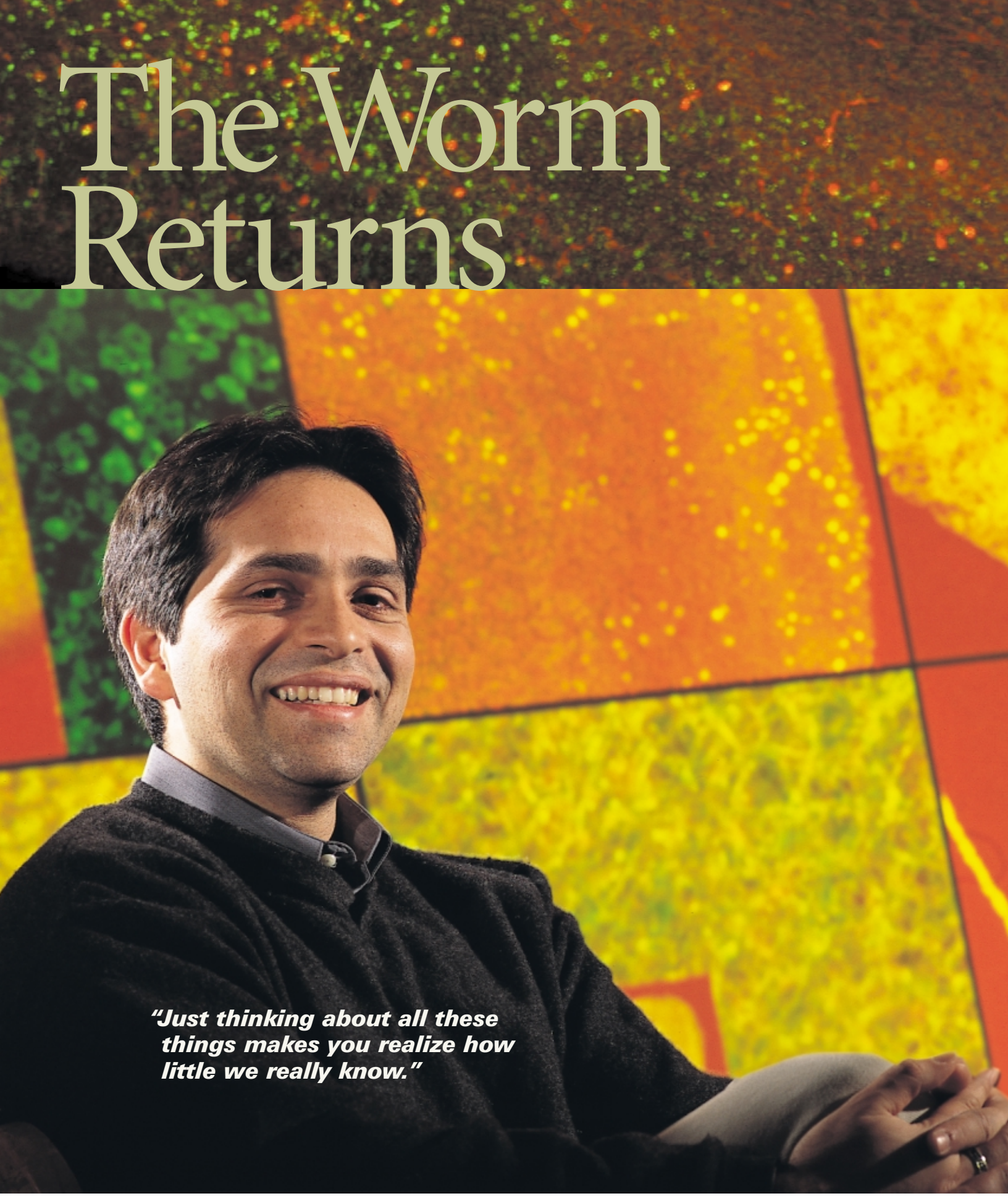
Alison Davis

Alison Davis

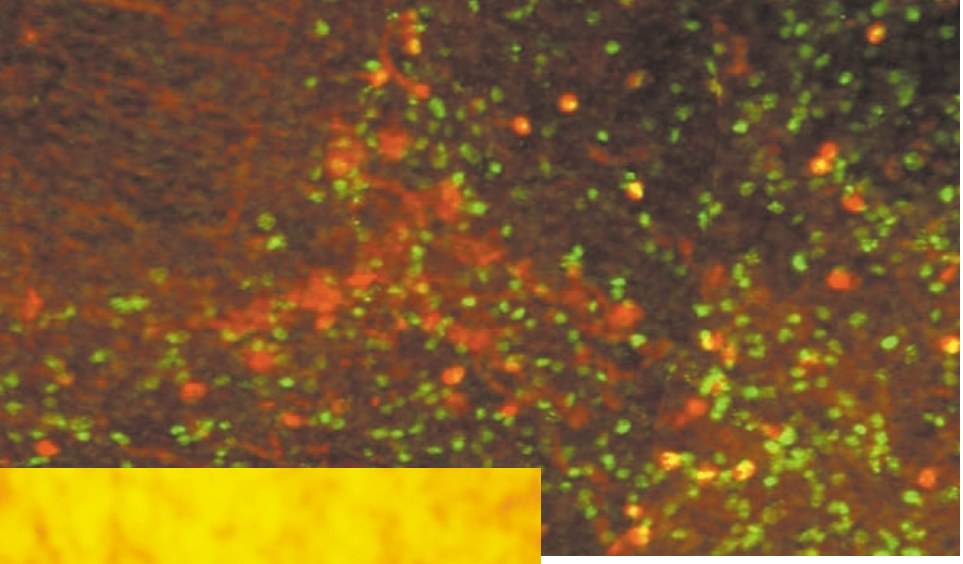
Editor

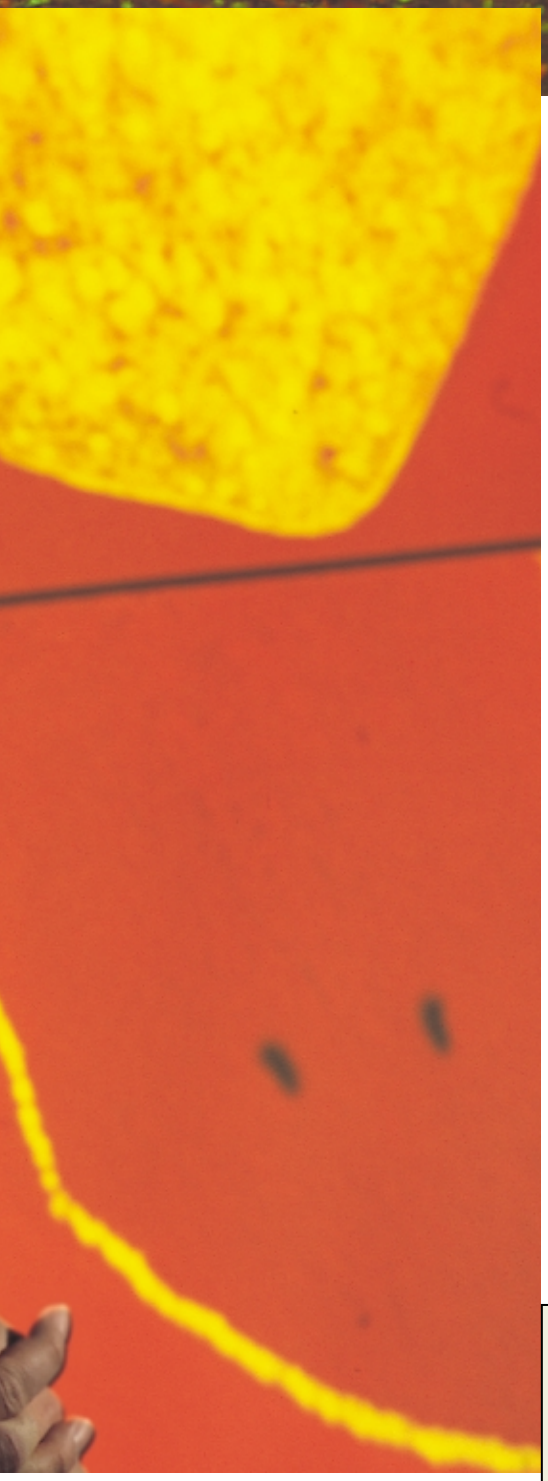
www.nigms.nih.gov/news/findings/feb01.pdf

The Worm Returns



"Just thinking about all these things makes you realize how little we really know."





William K. Geiger

by Alison Davis

He remembers the day like it was yesterday—not 21 years ago when that deciding moment actually took place.

Alejandro Sánchez Alvarado was sitting in his high school biology class, waiting to hear another round of facts from the teacher. And then, he recounts, "Professor Maldonado taught us how DNA copies itself."

"My teacher explained how Meselson and Stahl figured it out," says Sánchez Alvarado, referring to the two scientists who made this breakthrough discovery in the 1950s. They did it by ruling out two of three possible outcomes in a very simple experiment. "It just blew my mind to see how they used simple logic to figure it out," he remembers.

"That's what I want to do," he says he decided. He yearned to do hands-on biology experiments where "it's just you and the results."

After he completed high school in Caracas, Venezuela, at 17 years of age, Sánchez Alvarado headed northward to America, dead set on becoming a molecular biologist. But there remained a sticky problem: He spoke not a word of English.

So he moved to Nashville, Tennessee. "I knew nobody there spoke Spanish, so I'd have to learn English!" he recalls.

Sánchez Alvarado's pioneering spirit pushes forward today. The scientist, now 36 and a staff associate in the Department of Embryology at the Carnegie Institution of Washington in Baltimore, Maryland, is chipping away at a 250-year-old biological problem few researchers have taken on. Sánchez Alvarado wants to unravel Nature's secrets about how animals regenerate lost body parts after injury, and he's going about it using a tiny flatworm not many people in the world know much about.

An Amazing Little Worm

Meet *Schmidtea mediterranea*, an animal the size and shape of a toenail clipping, which is native to Barcelona, Spain. Commonly called a planarian, this aquatic organism is "truly amazing," says Sánchez Alvarado.

You don't have to be a scientist to agree with him: Anyone can cut these teeny worms up into many small pieces, and each piece "knows how" to make a new planarian of just the right size, with its head up and its tail down. How the pieces do that perfectly every time remains a mystery.

"This is what really keeps me awake at night," says Sánchez Alvarado.

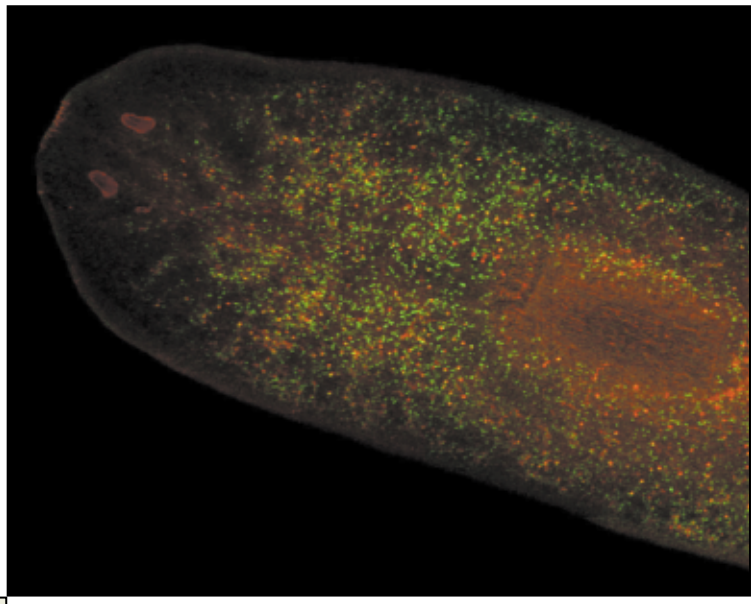
Alejandro Sánchez Alvarado is a biologist at the Carnegie Institution of Washington in Baltimore, Maryland.

Sánchez Alvarado admits to developing an insatiable curiosity about planarians after first learning about the ability of these organisms to entirely re-make themselves from a fragment a tiny fraction of the size of the original animal, an experiment performed in the late 1800s by the scientist Thomas Hunt Morgan.

"He did all kinds of cutting experiments—triangles, squares…he was cutting away like a madman," says Sánchez Alvarado, who keeps a copy of Morgan's classic 100-year-old article describing these experiments in detail close to his desktop computer.

"But planarians really stumped him."

According to Sánchez Alvarado, Morgan found it very puzzling that one small region of the worm (the area in front of its photoreceptors, or "eyes") simply would not regenerate, no matter what, even though under a microscope it looked similar, if not identical, to regions that could. Morgan eventually abandoned the study of planarians.



Phillip Newmark

Glow-in-the-dark fluorescent labels allow Sánchez Alvarado to peer into a microscope and see which cells in a flatworm have just copied their DNA and are getting ready to split in two.

Sánchez Alvarado and other scientists now know more pieces of the puzzle. Planarians, he says, have a stash of regenerative "stem cells" that are capable of roving to any wounded site, then turning into any of the approximately 30 different cell types that populate the adult worm. One conspicuous spot in a planarian where stem cells don't divide is—you got it—the region in front of its photoreceptors.

The challenge now, he says, is finding out *how* this happens. That is knowledge that may help scientists understand the intricate decisions cells make during development, and also after injury.

"It all boils down to one cell type," says Sánchez Alvarado, "the neoblast."

At the turn of the 20th century, all Morgan could do was to observe the neoblasts—the smallest planarian cell type that makes up 20 to 30 percent of its body—because the technology of the times didn't permit him (or anyone else, for that matter) to study their function.

Eventually, other scientists tried to overcome this impasse by using rather nonspecific techniques to mark the neoblasts so they could be seen under the lens of a microscope. The problem was that no one could figure out a way to trace the movements of these cells—a necessary task for understanding their seemingly magical ability to regenerate a cut-up animal.

Tracking the whereabouts and actions of neoblasts in planarians remained a sticky problem until quite recently, when Sánchez Alvarado and his postdoctoral fellow, Phillip Newmark, devised a way to detect newly copied DNA in neoblasts—a sign that those cells will soon split in two. The method enables Sánchez Alvarado and Newmark to track the regenerating cells as they divide and roam throughout the animal to create new body parts.

Keeping Things Simple

Scientific puzzles are huge and complicated, admits Sánchez Alvarado, but just like Meselson and Stahl's classic experiment, he thinks the best way to tackle them is to keep things simple.

Soon after his arrival at Carnegie, he focused hard on finding a model system to probe the mysteries of regeneration. Many of the other scientists "thought I was crazy," says Sánchez Alvarado. For the next few months, he didn't do a single experiment. Instead, he hopped on the commuter train to Washington, DC, roughly an hour's trek south, every day for 2 months and buried himself in the voluminous stacks of the biggest library in the world, the U.S. Library of Congress.

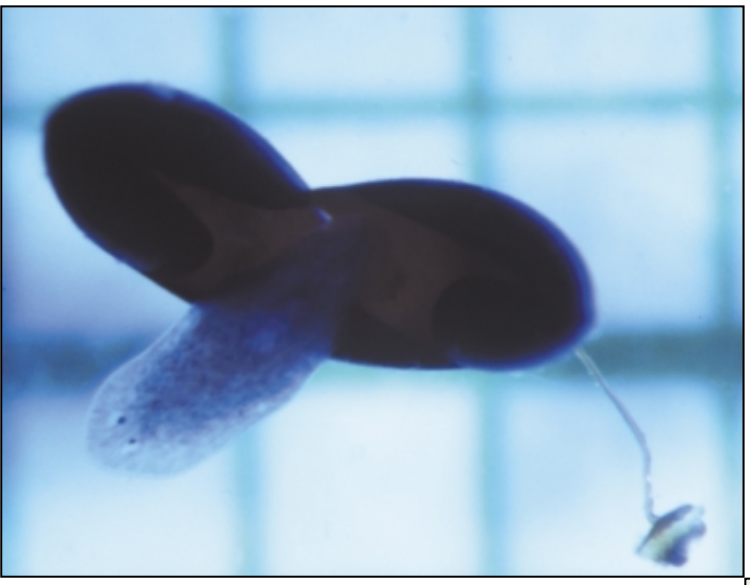
Sánchez Alvarado combed the stacks for 100-year-old publications on regeneration that existed nowhere else, and he learned that there is at least one organism from

every major branch of the biological kingdom that can regenerate itself. "Regeneration may have been present from the very beginnings of life," Sánchez Alvarado says.

People cannot grow back an arm, he says, but they do have tissues that regenerate constantly—the skin, the lining of the gut, and the liver, for instance. According to Sánchez Alvarado, scientists don't really know whether the molecular happenings that prod these tissues to grow back are the same as, or even similar to, those that cause a salamander to regrow a tail after it's been clipped off.

And how come a salamander with an amputated tail grows just a tail—and not a leg, or an arm, or even a whole back—Sánchez Alvarado wonders.

"Just thinking about all these things makes you realize how little we really know," says Sánchez Alvarado.



Alejandro Sánchez Alvarado

"These are very old, long-standing problems in biology, but they have been extremely difficult to study."

Finding an interesting and important problem is the first step, explains Sánchez Alvarado. But next, he adds, you must carefully choose the right system to help you solve the mystery. Regeneration researchers haven't studied planarians, he says, because the technical difficulties of working with them have been so frustrating, gaining these flatworms a reputation of being "impossible to work with."

Sánchez Alvarado thought hard about the problem and worked through many options. Hydras, for instance, are organisms even more primitive than planarians that regenerate body parts with ease. But hydras didn't cut it for Sánchez Alvarado: He ruled them out because these creatures are missing a primitive tissue type called "mesoderm" that gives rise to many human organs.

So studying hydras, Sánchez Alvarado explains, would not have allowed him to address questions about the regrowth of human tissues and organs, or about the way human embryos develop into adults.

Planarians, in contrast, do have mesoderm. And, perhaps more importantly to scientists like Sánchez Alvarado, they are consistently "diploid," meaning that each planarian cell contains an unchanging number of paired chromosomes. In contrast, the cells of many so-called model organisms contain varying numbers of chromosomes, making it virtually impossible for scientists to cross-breed them in order to study their genes.

"We need this kind of research to really understand human biology," says Sánchez Alvarado.

Obvious ethical limits prevent scientists like Sánchez Alvarado from studying people to address the kinds of questions that intrigue him and other molecular and developmental biologists. So instead, these researchers perform experiments with more primitive creatures—which, surprisingly, are endowed with a stunning array of the same genes we have inside every one of our cells.

Scientists often turn to model organisms like worms and flies to address fundamental questions about development and about tissue and organ formation. Many of the same genes controlling these molecular processes have been used over and over again throughout evolutionary time.

Starting From Scratch

The good thing about being one of the few people working on a problem is the abundance of unanswered questions. The bad thing is that the research tools a scientist needs to get going on such a project aren't necessarily readily available. So Sánchez Alvarado has had to make his own planarian toolkit, pretty much from scratch.

A hatchling of the planarian *Schmidtea mediterranea* bursts from its egg case.

In the science called genetics, the researchers' goal is to learn how the DNA inside every chromosome in all of our cells can create traits and influence behaviors. As with all organisms, from plants to people, the language of DNA (genes) spells out instructions for how to manufacture proteins, the molecules that do most of the work in our bodies. One way scientists go about figuring out what genes do is to get rid of them in model organisms and then see what happens.

To decipher how certain planarian genes encode signals to regrow a head or a tail, for instance, Sánchez Alvarado chose a similar approach.

He consulted a fellow Carnegie scientist, Andrew Fire, a developmental biologist who studies roundworms, a distant relative of flatworms. Recently, Fire pioneered a technique called "RNA interference," or RNAi, in which he feeds worms a meal laced with pieces of a cousin of DNA (called RNA) that can "erase" the activity of the worm gene Fire wants to delete.

With Fire's advice, Sánchez Alvarado got this method of eliminating genes to work in planarians.

To really be convinced that certain genes have been switched off, scientists like Sánchez Alvarado rely on another sort of technique, called *in situ* (literally, "in place") hybridization, in which genes that are "turned on" look purple or blue when viewed under a microscope. No color means the genes are either turned off or missing altogether. Researchers also use *in situ* hybridization to measure the activity of new genes they have engineered into test animals.

Sánchez Alvarado and members of his lab are also working on finding ways to identify cellular "landmarks" in planarians by testing dozens of antibodies that grip onto known cell structures in other organisms. Biologists who study individual cells often use such techniques to "label" parts of cells for reference purposes.

With lots of hard preparatory work in place, Sánchez Alvarado is ready for the next step.

Genes!

Sánchez Alvarado is searching for as many planarian genes as he can find, so he can put his newly minted tools to work and begin to crack the really tough problem of what they all do.

In particular, Sánchez Alvarado is interested in finding genes that affect regeneration that may shed light on the most primitive and fundamental questions about development. To date, he's snagged nearly 4,000—an amazing feat, since in addition to himself, only three other people work in his small lab.

Four thousand genes is a whole lot more than 25 genes, which is how many Sánchez Alvarado had unearthed a year before. That was before he embarked upon an extremely labor-intensive process of gathering tissue from hundreds of flatworms, extracting their DNA, and decoding the sequence—the order of building blocks called A, T, C, and G for short—of thousands of individual stretches of DNA (the genes). To accomplish this, Sánchez Alvarado enlisted the members of his small laboratory to join in an assembly line effort.

The end product of the assembly line is a series of DNA "letters"—hundreds of thousands of As, Ts, Cs, and Gs. Sánchez Alvarado is the one who takes care of the last step, spending hours at his computer sifting through the alphabet soup. He asks his computer to compare the newfound flatworm gene sequences to those of roundworms, mice, flies, and people, looking for matches. Since the function of many genes in these other organisms is already known, a sequence with lots of matches can sometimes unveil the planarian gene's function.

Chipping Away

But the dreams of gene hunters like Sánchez Alvarado do not end with finding the sequences of the genes. The next step involves learning what, when, and how the genes act, as well as how they work *together*. To do that, researchers often borrow tricks from computers, which are able to process lots of things at once. Scientists cram thousands of gene sequences onto a glass or plastic chip the size of a microscope slide and then, with the help of a computer, use this "gene chip" to scan the cellular activity of hundreds, if not thousands, of genes in one fell swoop. Such an approach can tell a scientist which genes get switched on during wound healing or in a certain type of infection, for example.

The chips could also reveal gene activity as regeneration takes place.

Sánchez Alvarado plans to custom-design a planarian gene chip himself. He is not the slightest bit intimidated by all the work involved in setting up such a system, because he believes it will deliver mounds of interesting information.

"It's hard, but that's why I like this so much," he says with an unquestionable zeal.

Information is already pouring out.

The results have been astonishing, says Sánchez Alvarado. Many of the genes revealing themselves are extremely similar to human genes, fanning Sánchez Alvarado's fire to apply the knowledge to health-related matters, such as wound healing.

But another fallout from the work may be its potential impact on scientists' understanding of phylogeny, the study of how organisms are related to each other on the evolutionary tree.

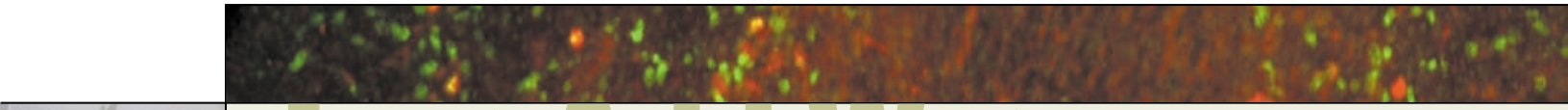
"Nobody seems to agree on the phylogenetic tree for [many-celled animals]," says Sánchez Alvarado. The problem, he explains, is that most of the distinctions defining which branch goes where have been based purely on either looks—in scientists' terms, "morphology"—or on limited gene sequencing data.

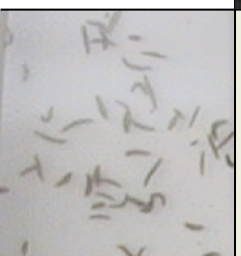
To make matters worse, when researchers take into account both morphology and DNA sequence to build phylogenetic trees, they find more and more examples that "break the rules." As it turns out, a creature that looks most like another is not necessarily closely related to it. For example, Sánchez Alvarado is finding many genes in planarians that people have, but fruit flies don't. That shouldn't be possible according to the way current evolutionary trees are drawn, in which planarians came first, then flies, then people.

Who cares? Well, scientists use phylogenetic assignments all the time to assemble pieces of facts into a body of knowledge—about how limbs first evolved, for instance. A deep understanding of these issues will be vital to help scientists get a grip on how to diagnose, and even fix, birth defects and other developmental disorders.

Sánchez Alvarado hopes that in the future, his work may contribute to other health benefits. He wonders if it might be possible someday to create a regeneration program in people with amputated limbs, for example. Time, and thousands more experiments, will tell.

"There are lots of things we still need to know in order to really understand ourselves," he says.





Just Add Water

Planarians are *so* predictable. Snip them any which way, and voila! In a couple of days, a worm of the right size and shape emerges.

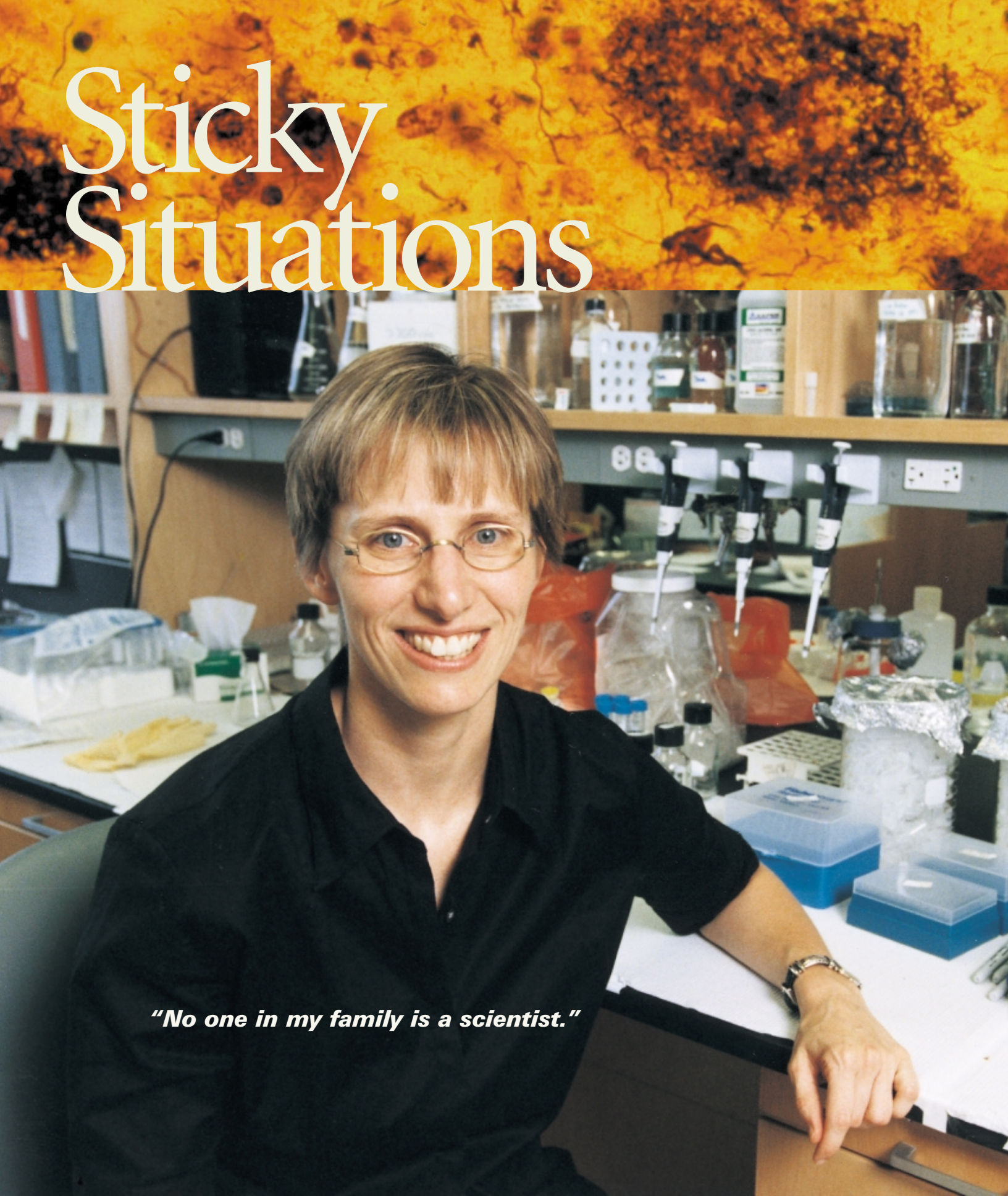
But despite the fact that these animals don't require extraordinary amounts of care and feeding, when Alejandro Sánchez Alvarado and his postdoctoral fellow Phillip Newmark first introduced planarians to their lab, they failed miserably. Left and right, the worms died. Sánchez Alvarado discovered the hard way that the Tupperware® homes they had set up for the planarians were "luxury condominiums," he says. "In the wild, they live in dirty water. Ours was way too clean." Supplementing the water with loads of minerals and salts eventually made the worms thrive.

But really, planarians are incredibly easy to keep, Sánchez Alvarado says. He spends all of $5 per year on grocery-store liver to make a paste to feed his planarian colony, and they live happily in clear plastic bins in a chamber kept at about 65 to 70°F (provided the water isn't too clean!). Nutrition and temperature are the only things that impact the growth of these flatworms. Cut back on their food and they shrink a bit; feed them too much and they just split in two. When Sánchez Alvarado traveled to a meeting in Europe last summer, he stashed them for a month in a refrigerator set to 46°F. They were fine.

Schmidtea mediterranea, the species of planarians Sánchez Alvarado works with, have an interesting place of origin: a filthy, old fountain in downtown Barcelona, Spain (see photo). Newmark was the one who first learned of this species of worms while working in Spain a few years ago. However, in general, planarians are aquatic animals, and they are abundant in a variety of fresh-water environments, such as underground aquifers and rivers. —*A.D.*

Phillip Newmark and Alejandro Sánchez Alvarado

Sticky Situations



"No one in my family is a scientist."

Neil Kowall and Ann McKee

Jeff Miller

Laura Kiessling (left) is a chemist at the University of Wisconsin, Madison.

The brains of people with Alzheimer's disease are riddled with abnormal proteins and tangled fibers (top).

by Alisa Zapp Machalek

Laura Kiessling started thinking and acting like a scientist before she really knew what one was. "No one in my family is a scientist," explains the University of Wisconsin, Madison chemist. "But my brother Bill and I used to like to do experiments when we were little." The siblings started out with a standard experiment kit that included instructions for making a lie detector and other gadgets. Once they'd exhausted all the kit's standard uses, the siblings got creative. "We rigged it up to give a small shock to the doorknob and then tricked our younger brother Mark into opening the door."

Kiessling, 40, now does experiments on a more serious topic: inflammation. She explains her work this way: "Say you smash your finger and it swells up. That's because white blood cells have been recruited to that area. One of the big questions to ask is: How do they know where to go?"

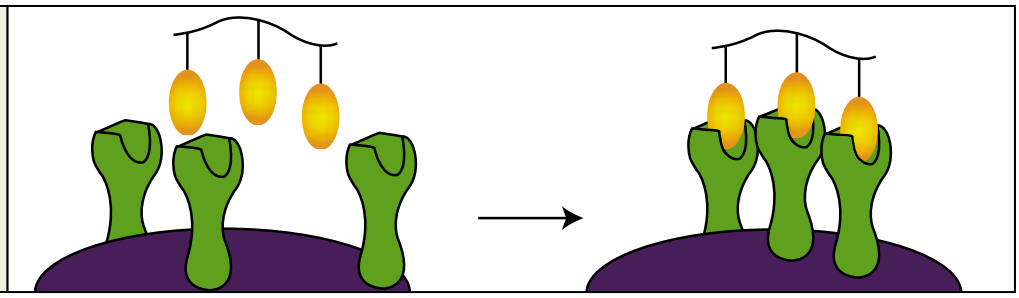
This question grows from Kiessling's main research interest, which is in understanding how cells stick to each other. The ability of cells to adhere selectively to other cells is key not only to inflammation, but also to a host of other biological processes ranging from fertilization, in which a sperm cell sticks to an egg, to bacterial infection, in which the bacteria must attach to host cells before infecting them.

Sweet-Talking Cells

Cell stickiness, or "adhesion," is made possible by proteins and sugar molecules that stud the surfaces of cells. Far more than just a candy coating, these proteins and sugars move around in the cell's outer membrane and grab onto molecules on the surface of other cells. Kiessling's studies are revealing how some of these sticky molecules enable cells to adhere to each other.

Long strings of sugar molecules have a familiar name: carbohydrates. When most people hear the word carbohydrate, they probably think of pasta, fad diets, and food labels. But carbohydrates are also the key players in a huge variety of biological processes. Immune cells protect us by recognizing and reacting to carbohydrates on the surface of bacteria and viruses. Cancers can spread throughout our bodies by using their cell-surface carbohydrate molecules to latch onto healthy cells.

A key to inflammation, and a focus of Kiessling's studies, are molecules called L-selectins. These molecules are proteins that sit on the surfaces of white blood cells, immune system cells that help fight infection and, when overactive, can cause inflammation.



Kiessling's tailor-made molecules (orange) force L-selectin molecules (green) to cluster together on the surface of a white blood cell. Kiessling is examining whether such molecules could be used to treat inflammation.

Laura Kiessling

To respond to an injury, white blood cells must slip out of the flowing bloodstream and move into the damaged tissue. But first, they have to slow down and stick to the wall of the blood vessel. That's where L-selectin molecules come in. They act as white blood cells' brakes by latching onto molecules that protrude from the blood vessel wall.

L-selectins are rather picky about which molecules they grab. They favor glycoproteins, which are proteins crowned with sugars. Like a molecular sweet-tooth, L-selectins grasp onto these sugars. Glycoproteins encourage the interaction by displaying many copies of their sugar groups for L-selectins to grasp.

To get a better handle on this process, Kiessling designed molecules to mimic glycoproteins. In essence, she created sugary decoys to trick L-selectins. When L-selectins bind to these decoys, they can no longer stick to their natural glycoprotein partners.

Kiessling knew she had to make her molecular mimics extra sweet to divert L-selectins from these natural partners. So she designed decoys that contain a whole string of sticky sugar groups that are nearly irresistible to L-selectins. In fact, L-selectins actually rearrange themselves in the white blood cell membrane in an attempt to grasp every possible sugar group.

Kiessling calls her decoys "multivalent ligands" because their many sugar groups provide multiple binding sites for L-selectins. "Ligand" is a generic term for a small molecule that binds to another, usually larger, molecule.

Kiessling's ultimate goal is to be able to control the behavior of cells. By using different multivalent ligands, she can direct the way L-selectins arrange themselves into clusters. For instance, a ligand with five sugar groups (five binding sites for L-selectin) will force L-selectin molecules to group themselves into clusters of five in the cell membrane.

"The way proteins are organized on a cell affects how that cell works," says Kiessling. "With our chemistry, we can make ligands that cluster proteins in the cell membrane to different extents. By varying how many binding groups we display, we can make clusters of three proteins or five proteins, and see how this influences the cell's response."

Reining in the Immune Response

Normally, soon after L-selectins cluster together in a cell membrane, they are clipped off the cell surface. In the presence of multivalent ligands, and without their L-selectin grappling hooks, white blood cells are unable to stick to the blood vessel wall and can't participate in an immune response. So this clipping process could protect the body against excess inflammation, says Kiessling.

"If you could trigger the release of L-selectin, you could strip it off cells that would normally get recruited in inflammation so that they wouldn't be recruited," she reasons. With fewer cells involved, the immune response might be reduced. "That would be good if you were trying to develop new anti-inflammatory strategies."

Kiessling is working on just that. She knows her multivalent ligands force L-selectin molecules to cluster on the membrane of white blood cells. She'd like to learn whether they also prompt the release of L-selectin—and, as a result, could reduce inflammation. So she is working with other scientists to test her ligands as anti-inflammatory agents in animals.

Kiessling is also studying how protein clustering affects the production of antibodies, which are molecules that attack foreign substances like bacteria and viruses. By slightly varying the design of a multivalent ligand, Kiessling can force the immune cells that make antibodies, called B-cells, to start or stop churning out the molecules.

Controlling antibody production could have many practical applications, Kiessling predicts. "We might be able to make a potent vaccine by having ligands that activate antibody production." On the other hand, she says, ligands that prevent antibody production may help researchers treat autoimmune diseases or suppress a body's rejection of a transplanted organ.

Untangling Alzheimer's Disease

While one secret to treating inflammation may be to force proteins to cluster together, for other diseases the key may be to keep proteins apart. Using the flip side of her L-selectin clustering strategy, Kiessling is trying to find a way to untangle clusters of abnormal proteins that accumulate in the brains of people with Alzheimer's disease.

Alzheimer's disease is a progressive, incurable disorder that usually affects people over 65. It impairs memory and thinking as brain cells die.

One of the reasons Alzheimer's disease is so difficult to treat is that scientists don't really know its root cause. Many suspect that one culprit is an abnormal form of a protein called beta-amyloid that clumps together into tangled fibers in the brains of people with the disease.

Kiessling and her collaborator Regina Murphy, a chemical engineer at the University of Wisconsin, Madison, reasoned that if they could disrupt the formation of beta-amyloid fibers, they might be able to stop the progression of the disease. Together, they created ligands designed to bind to beta-amyloid and prevent it from clumping together. These so-called "monovalent" ligands each bind to only one molecule of beta-amyloid and are designed to break up the protein clumps.

The ligands actually work on the same principle as everyday detergents. The molecules have one half that grabs the dirt, grease, or other goop—in this case, clumps of beta-amyloid proteins. The other ligand half forces the goop to dissolve in water, even helping to break up the clumps.

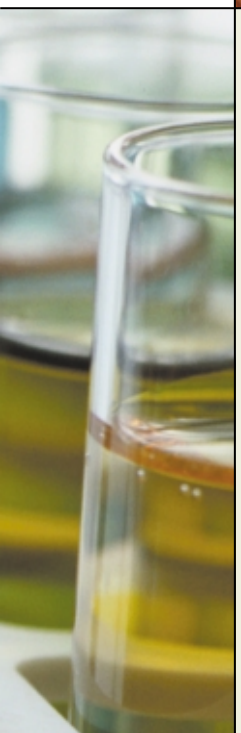
The good news is that the ligands do prevent beta-amyloid clusters from killing nerve cells in a plastic dish. The perplexing news is that they don't work the way Kiessling and Murphy expected they would. "These compounds were designed to block protein aggregation [clumping], but they don't block aggregation," says Kiessling. "Instead, they change the structures of the aggregate."

Kiessling discovered that after exposure to the monovalent ligand, the clusters of beta-amyloid were not long fibrils that piled up in a solid mass inside brain cells. Instead, they were shorter, more highly branched structures. The finding could change the way scientists think about the molecular causes of Alzheimer's disease.

According to Kiessling, it may also mean that you don't necessarily need to completely block protein clumping to have a useful drug to treat Alzheimer's disease.

"Everyone with Alzheimer's presumably already has aggregated beta-amyloid in their brain, so if you needed to stop aggregation, your compound would have to be taken [before protein clumping even started], which is not very attractive or practical," Kiessling explains.

What's more, she says, you'd have to take enough of the drug to bind up every beta-amyloid protein. But if all

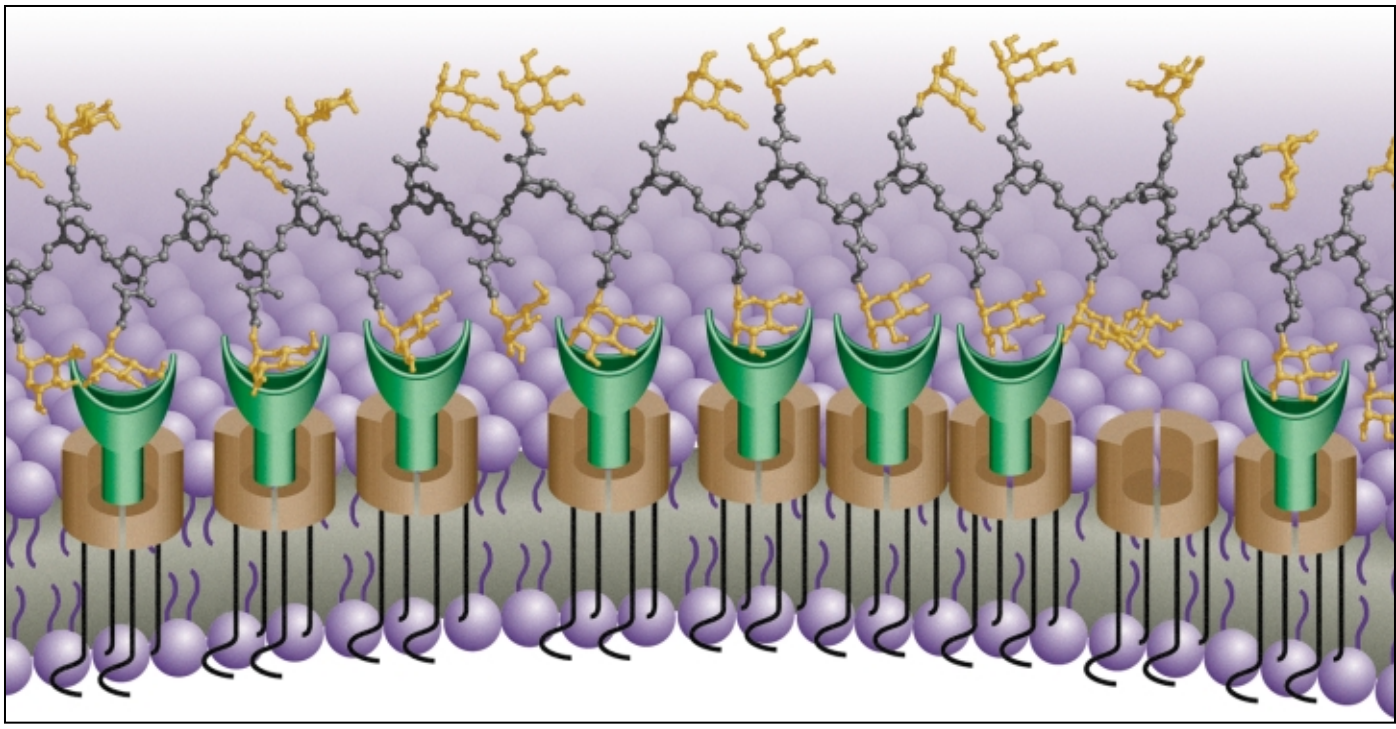


Genius at Work

Some people call Laura Kiessling a genius. At least the MacArthur Foundation—which issues the so-called "genius awards"—has done so. In 1999, Kiessling won this prestigious award for using chemistry to improve our understanding of the biology of inflammation.

Actually, the MacArthur Foundation admits the award's nickname is a misnomer, because the foundation designed the fellowship to recognize not only intelligence, but also exceptional creativity and promise. In Kiessling's case, this includes the ability to make connections between seemingly unrelated scientific fields. Her studies reveal how molecules assemble and may have implications for understanding, and possibly treating, conditions such as Alzheimer's disease and organ rejection.

The MacArthur Fellowship gives Kiessling $285,000 over a 5-year period to spend any way she would like. The fellowships are "no strings attached" cash awards designed to encourage talented and creative individuals in virtually any field. Unlike most awards, the MacArthur Fellowship does not require its awardees to write status reports or to work on a specific project. Recipients, who number 20 to 40 each year, include writers, composers, scientists, teachers, humanists, activists, poets, and many others. —*A.Z.M.*



Laura Kiessling

you need to do is disrupt further aggregation, as her studies suggest, you just need a small amount of a drug.

In addition to revealing a potential treatment strategy for Alzheimer's disease, Kiessling's work could improve scientists' understanding of other diseases that involve protein clumping, such as "mad cow" disease and possibly Lou Gehrig's disease, also known as ALS.

Unlikely Connections

Although Kiessling now studies biological problems like inflammation and Alzheimer's disease, she was originally trained as an organic chemist. She earned a bachelor's degree from the Massachusetts Institute of Technology and a doctorate from Yale University, both in chemistry. She has been on the chemistry faculty at the University of Wisconsin, Madison since 1991, and she joined the biochemistry faculty there in 1997.

"It's very significant that she's moved so far from her formal training," says John Schwab, a chemist at the National Institute of General Medical Sciences, which funds Kiessling's research. "As a chemist, Kiessling is beautifully trained to take advantage of techniques that might be unrelated to physiology [living systems] and apply them in really imaginative ways to the study of complicated biological processes. A good example of that is her use of ROMP technology."

ROMP stands for ring-opening metathesis polymerization. Like other polymerization reactions, ROMP creates long chains of identical molecules. The ROMP chemical reaction was originally developed by Robert Grubbs, a chemist at the California Institute of Technology. Most scientists use the technique to make chemicals with useful mechanical, electronic, or optical properties. But Kiessling adapted the method to string together sugars and create a wide variety of carbohydrate ligands that bind specifically to molecules on the surfaces of cells.

Carbohydrates are some of the most difficult biological molecules to study. In the past few decades, scientists have streamlined techniques to tailor-make proteins, but making customized carbohydrate molecules is not so easy. Recent advances, including Kiessling's use of ROMP technology, have helped even the playing field.

Using ROMP, Kiessling can customize the number, density, and arrangement of binding sites on her ligands. She can even track or immobilize the ligands by adding special chemical tags to their ends. These tailor-made ligands allow Kiessling to control the signals cells receive—and the cells' responses.

Joining Chemistry and Biochemistry

Kiessling not only studies stickiness, she creates a bit of her own. She's joined her two labs in a cohesive project,

with her chemistry students making the molecules and her biochemistry students studying how the molecules work in living cells.

"What I like is that the chemistry students really get a good understanding of the biochemistry experiments and the biochemistry students come away with a much better understanding of what compounds are possible to make," says Kiessling. "The collaboration is very tight. On a daily basis, my students aren't only talking to me, but the chemists and the biochemists are talking to each other."

Kiessling can also sympathize when her students' experiments don't work out as planned. As with any experiment, there are bound to be failures. Kiessling even remembers some from her childhood days.

"One time I tried to put little motors on the end of a Frisbee that would fire in opposite directions to make it spin really fast," she recalls. The Frisbee wouldn't fly at all, but instead ended up sputtering and convulsing on the lawn. "I forgot about the idea of buoyancy," Kiessling says. "Let's just say it wasn't very aerodynamic."

To study conditions and processes ranging from inflammation to Alzheimer's disease to bacterial movement, Kiessling creates long, sticky carbohydrate molecules (orange and gray) that interact with receptors (green) on the surfaces of cells.

These days, Kiessling tries to pass on her excitement and enthusiasm about chemistry to her students. "I really enjoy teaching," she says. "And I have some great stories about it."

One of her favorites is when she was teaching a class about chemical reactions. The big point she was trying to convey, she recalls, is that a catalyst (a molecule that facilitates a chemical reaction without being changed in the process) can't change the equilibrium, or balance, of a chemical reaction. It just reduces the amount of energy required for the reaction to occur.

"I happened to be reading *The New York Times* magazine and saw an ad for the perfume Catalyst. It showed this woman in a long black, strapless evening gown standing in the middle of the prairie with three cowboys leaning over a fence leering at her. The caption said: 'Catalyst: Disturb the equilibrium.' I made a slide of it and asked my students to come up with a chemically correct slogan for the perfume. The students came up with a whole bunch of great answers—some of which are definitely not fit to print."

Kiessling's work continues to shed light on the chemistry behind cell stickiness—and a host of associated diseases. In recognition of the significance of her work, she has already received numerous honors and awards. These range from being one of the first two winners of a Distinguished Alumni award from her high school in Wisconsin to receiving the MacArthur Foundation "genius" award (see sidebar).

Ron Raines

Kiessling greets her daughter, Kyra, after returning from her first trip away—to review research grant applications for the National Institutes of Health.

One of Kiessling's biggest fans has an office in the same building. Her husband, Ron Raines, is a biochemist at the University of Wisconsin, Madison. While Kiessling focuses on carbohydrates, Raines studies proteins and seeks to understand how a protein's shape relates to its role in the body.

The two work together on some scientific projects, but their most cherished partnership—as parents—transcends research.

The couple now have a young daughter, Kyra, whose name means shining light and was derived by combining the last names of her parents (Kie-Ra).

In Kiessling's eyes, her roles as scientist and parent compliment each other perfectly.

"Many people don't know what it's like to be an academic scientist—how flexible and multifaceted your job is. You never know for sure what you'll end up doing that day. It's the perfect preparation for parenthood."

"Besides," she continues, "children are really like little scientists. They're always experimenting. And that's what scientists do."

Cancer Patients Breathe Easier After This Test

NIGMS grantee **Paul Watkins** at the University of North Carolina at Chapel Hill has come up with a simple breath test that measures an individual cancer patient's ability to break down a medicine called docetaxel. The test measures the strength of a protein nicknamed "CYP3A4" that chews up docetaxel as it passes through the liver. The CYP3A4 protein processes many different drugs, including the antibiotic erythromycin. To administer the test, Watkins injects a tiny amount of erythromycin containing trace levels of a radioisotope of carbon, then measures the amount of radioactive carbon dioxide the patient exhales 20 minutes later. (The very small amount of radioactivity used poses no danger to the patient or the health care provider.) Underscoring the value of this simple test in helping doctors find the best docetaxel dose, Watkins and his coworkers found that the patients with the lowest scores on the breath test were the ones who suffered the greatest toxicity from this cancer treatment.

Souped-Up Artificial Skin Heals Burn Wounds

Twenty years ago, burns covering half the body were routinely fatal. Today, patients with burns encompassing 90 percent of their body surface can survive, albeit sometimes with permanent impairments. Survival statistics have risen sharply over this time in part due to insights stemming from basic research aimed at understanding how skin responds to burns. NIGMS grantee **Steven Boyce** at the University of Cincinnati and the Cincinnati Shriner's Burns Hospital has recently figured out a way to grow skin cells from a burn patient and then add them to a polymer sheet to create living skin grafts in the lab. To try to permanently close burn wounds, Boyce placed the lab-grown skin grafts on top of an artificial skin called Integra™ and bathed everything with a nutritious mix of growth factors and antibiotics to help prod the growth of new blood vessels and control infection. The technique was tested on three children who had been badly burned in fires. In each case, the patient's new skin was a lighter color than before, but it returned to its original softness, smoothness, and strength with minimal scarring.

A Little Fish With Your Aspirin?

Aspirin is one of the oldest medicines around, but believe it or not, scientists do not know exactly how it can do so many things, from treating sunburns to preventing heart disease. The basics are clear—aspirin blocks an enzyme in our bodies called cyclooxygenase (COX). But new facts are emerging as scientists ponder how medicines interact with each other and with other substances, such as foods. NIGMS grantee **Charles Serhan** at Brigham and Women's Hospital in Boston, Massachusetts has found that aspirin and certain fish oils (omega-3 fatty acids abundant in salmon and other fish) can work together to fight painful inflammation and blood vessel disease. Inflammation is a normal process; it is an important way our body summons immune cells to fight infections. But sometimes, the immune system overreacts, sending too many white blood cells to an infection site. There, the overcrowded cells spill their toxic, disease-fighting arsenal into the blood, triggering nerve cells to fire. This can cause pain. Serhan and his research team have discovered that the same cellular enzyme that aspirin interacts with can help inflamed white blood cells to manufacture their own anti-inflammatory chemicals from molecules in fish oil. This can help aspirin produce even more of its beneficial effect.

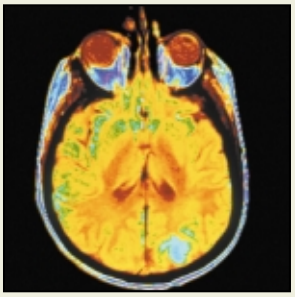
Bacterial Slime a Hallmark of Cystic Fibrosis Lungs

Thick mucus clogs the lungs and intestines of the approximately 30,000 Americans* who have cystic fibrosis (CF). This accumulation of mucus leads to malnutrition, frequent airway infections, difficulty breathing, and permanent lung damage. Bacteria thrive in the nutrient-rich mucus, and tragically, most patients with the disease die from overwhelming lung infections. One bacterium in particular, *Pseudomonas aeruginosa*, is the culprit in most of these lethal infections. Recent research conducted by NIGMS grantee **E. Peter Greenberg** at the University of Iowa concludes that to make matters worse, this microbe gains a nearly unbreakable foothold in CF lungs by forming organized groups encased in a self-produced slime, called a biofilm. Biofilms coat a variety of surfaces and account for everything from dental plaque to unsightly toilet bowl stains. But in our bodies, biofilms signify serious trouble, as they are stubbornly resistant to antibiotic treatment. Greenberg and his colleagues developed a sensitive lab test that detects telltale biofilm-manufactured molecules. The researchers expect that the test will speed the discovery of new drugs to combat biofilms.

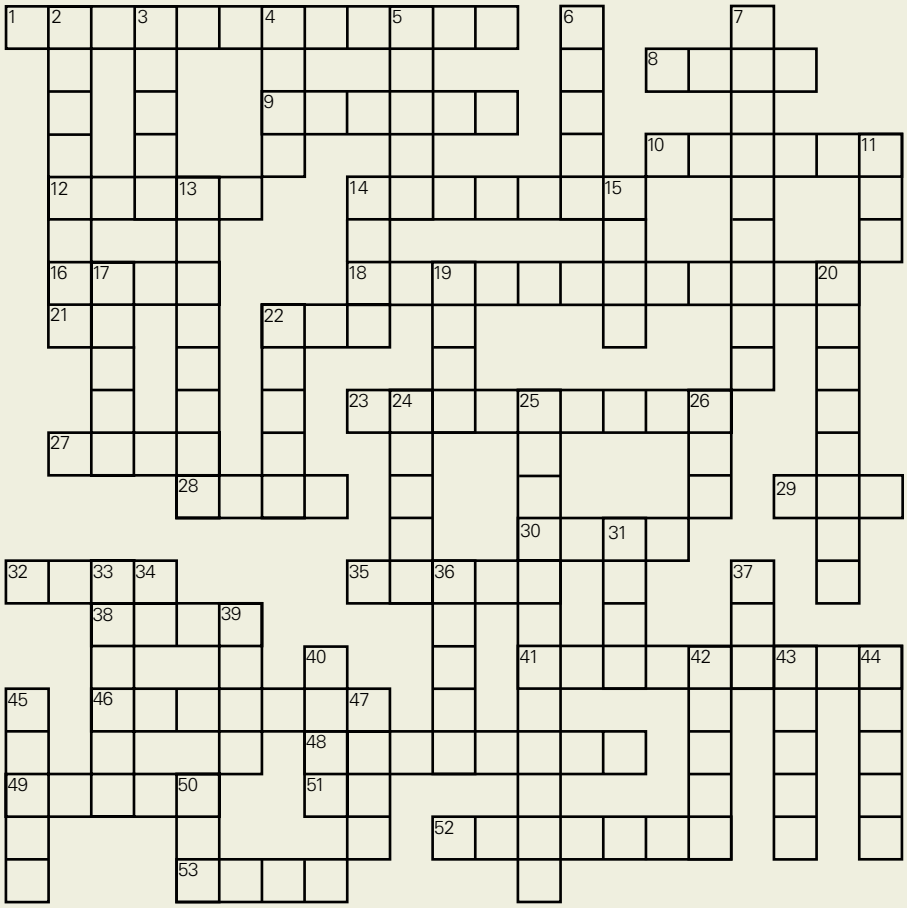
*According to data from the Cystic Fibrosis Foundation

Seeing Clearly Now

Physicians routinely use a technique called magnetic resonance imaging (MRI) to diagnose disease. For example, the method helps doctors find tumors in the body that are impossible to detect by other means, such as X-rays. MRI works by finding, with the help of a strong magnetic field, water molecules scattered throughout the body. Since water is the main constituent inside all of our cells, finding it "paints a picture" of what's underneath our skin. To help distinguish between different types of tissues—or between healthy cells and tumors—radiologists often inject liquids called contrast agents into a patient's bloodstream just before an MRI procedure. These dyes spread through the blood and interact with water molecules exiting cells. This process occurs differently in healthy and diseased cells, which accounts for the contrast dyes' ability to detect tumors. But the rate at which MRI contrast dyes interact with water isn't exactly what scientists had assumed, says NIGMS grantee **Charles Springer** at Brookhaven National Laboratory in Upton, New York. This can cause significant errors in interpreting the results, he says. Springer's recent findings promise to improve the ability of contrast dyes to more accurately label tumors and other types of diseased tissues.



The Last Word



ACROSS

1. scientific name for sugar
8. basic constituent of any living organism
9. nickname for Laura Kiessling's award
10. kicked
12. sweet stuff
14. in the lead
16. leave out
18. lost body parts regrown
21. not yes
22. edge
23. the science of studying chemicals
27. looked at
28. useful for knitting and weaving
29. where scientists work
30. breathing organ
32. seen atop an angel's head
35. simple aquatic creature and mythological water serpent
38. contains eye color
41. drugs
46. pill for a headache
48. molecular "brake" for white blood cells
49. kind of organism used in the lab
51. Santa's first word
52. slimy bacterial community
53. big quiz

Puzzle answers can be found at www.nigms.nih.gov/news/news.html

DOWN

2. cell stickiness
3. carry
4. relaxation exercise
5. excuse
6. tearful vegetable
7. type of flatworm
11. gene letters
13. immune system protein that fights invaders
14. what the early bird gets
15. microbe
17. bucks
19. protein-making instructions
20. primitive cell that can create a planarian
22. ship's hearing
24. not sad
25. immune reaction causing swelling and pain
26. not no
31. require
33. molecule binder
34. not and, not but
36. took the car
37. med. imag. scan
39. body covering
40. lots of them in the sea
42. after ice
43. basic research supporter
44. to do before a test
45. Haley has one
47. makes a sign bright
50. aflame

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