Excerpt from the June 11, 1966 issue of *Science News*

50 YEARS AGO

3-D Home TV Foreseen

The pace of new developments in the recently revived method of photography known as holography is so fast that three-dimensional television sets portraying life-size scenes could be a reality before 1984, as was predicted in George Orwell's novel.... A hologram is a recording of an interference pattern reflected from an object. From this recording, the object can be reconstructed visually in a three-dimensional form.

UPDATE: Television viewers are still waiting for the 3-D revolution. Although 3-D TVs went on sale in the United States and elsewhere in 2010, they have yet to take off. Most sets require special glasses or have limited viewing angles, and none use holography to create the illusion of depth. Scientists haven't given up, though. Using innovative plastic screens, researchers are projecting small holographic movies in real time (*SN: 12/17/11, p. 18*). The enormous bandwidth and processing power needed to transmit and display the images are still huge barriers to making Orwell's vision a reality.

Kate Rubins will put her years as a molecular biologist and her astronaut training to good use when she rockets to the International Space Station on June 24.

THE SCIENCE LIFE

Space dreams get real

When molecular biologist Kate Rubins blasts off from Kazakhstan on June 24, strapped into the Soyuz spacecraft bound for the International Space Station, the trip will cap off seven years of preparing — and 30 years of hoping.

As a child, Rubins plastered her Napa, Calif., bedroom with pictures of the space shuttle, proudly announcing her intention to be an astronaut. A week at Space Camp in Huntsville, Ala., in seventh grade cemented her vision. But by high school, she concluded that astronaut wasn't "a realistic job," she says.

Flash forward to 2009: Rubins is running a lab at the Whitehead Institute for Biomedical Research in Cambridge, Mass., focusing on virus-host interactions and viral genomics. A friend points out a NASA ad seeking astronaut candidates, and Rubins' long-dormant obsession awakens. Since then, she has learned how to fly a T-38 jet, speak Russian to communicate with her cosmonaut crewmates, conduct a spacewalk, operate the robotic arm on the ISS and even fix the habitable satellite's toilet.

Joining NASA meant leaving her 14-person lab behind. But Rubins gained the rare opportunity to collaborate with dozens of scientists in fields as diverse as cell biology and astrophysics. On the space station, she'll be "their hands, eyes and ears," conducting about 100 experiments over five months.

She will, for instance, probe how heart cells behave when gravity doesn't get in the way. And she'll test a hand-held DNA sequencer, which reads out the genetic information stored in DNA and will be important to future missions looking for signatures of life on Mars.

At times, Rubins will be both experimenter and subject. In one study, she will observe bone cells in a lab dish, comparing their behavior with what happens in a simulated gravity-free environment on the ground. Because astronauts in space are vulnerable to rapid bone loss, CT scanning before and after the mission will also document changes in Rubins' own hip bone.

Rubins is particularly eager to examine how liquid behaves in microgravity on a molecular scale. In 2013, Canadian astronaut Chris Hadfield created an Internet sensation when he demonstrated that wringing out a wet washcloth in space caused water to form a bubble that enveloped the cloth and his hands. "It's incredibly bizarre," Rubins says. Understanding how fluids move in test tubes in space will help NASA plan for Mars exploration, among other applications.

Before any of the research can begin, Rubins has to get off the ground. As treacherous as accelerating to 17,500 miles per hour may sound, she's not worried.

"An important part of the training experience is making all the information and skills routine," she says. She predicts that sitting down in the Soyuz spacecraft, pulling out her procedures and getting ready to launch will feel a lot like going into the lab and picking up a pipette — "a normal day at the office."

Until the engines turn on, anyway. "I think it's going to feel different when there's a rocket underneath."

— *Siri Carpenter*

ATOM & COSMOS

Aging moon mission still going strong

After 7 years, NASA orbiter continues to discover lunar secrets

BY CHRISTOPHER CROCKETT

Not all cosmic mysteries lie light-years away. Some secrets are being unearthed on our nearest neighbor, about a quarter of a million miles from home.

For almost seven years, NASA's Lunar Reconnaissance Orbiter has been keeping a close eye on the moon. During its tenure, the spacecraft has cataloged craters, pinpointed subsurface deposits of water ice and found evidence of recent volcanic activity. It has even witnessed crashes by three other spacecraft.

"No other mission has orbited the moon for as long as LRO has," says geologist Noah Petro of NASA's Goddard Space Flight Center in Greenbelt, Md. Constant lunar vigilance has "really pushed our understanding of how the moon changes today, over the last billion years and what happened early on." A July 15 special issue of *Icarus* celebrates the mission's many discoveries, which flesh out not only the moon's story, but also reveal how Earth and the other rocky planets have been pummeled by space debris over the last 4 billion or so years.

When LRO launched on June 18, 2009, its goals were more modest. The spacecraft was sent to scout landing sites for future astronaut expeditions, hunt for resources such as water and better understand the radiation hazards that human crews traveling to the moon would face. Since completing its original one-year assignment, the mission has been extended several times. LRO plans to stay busy through September, and the LRO team has asked NASA for two more years.

Water ice turned up in some unexpected places. Other spacecraft had previously seen hints of water, but none could map precisely where it was. Researchers suspected that water lay within permanently shadowed craters at the poles, and LRO did find evidence of ice there. But LRO also found that not all shady spots harbor water, and not all water is found in the shadows — some appears to hide under soil that sits in direct sunlight.

"That was a bit of a surprise," says LRO project scientist John Keller, also at Goddard. Looking at temperature alone, it seems, isn't enough for understanding the history of water on the moon. In the polar shadows, where temperatures hover around –250° Celsius, water ice can endure for billions of years. But elsewhere, water may have been trapped more recently and protected by the terrain. "There's an interplay with time, temperature and topography underlying this water story," Keller says.

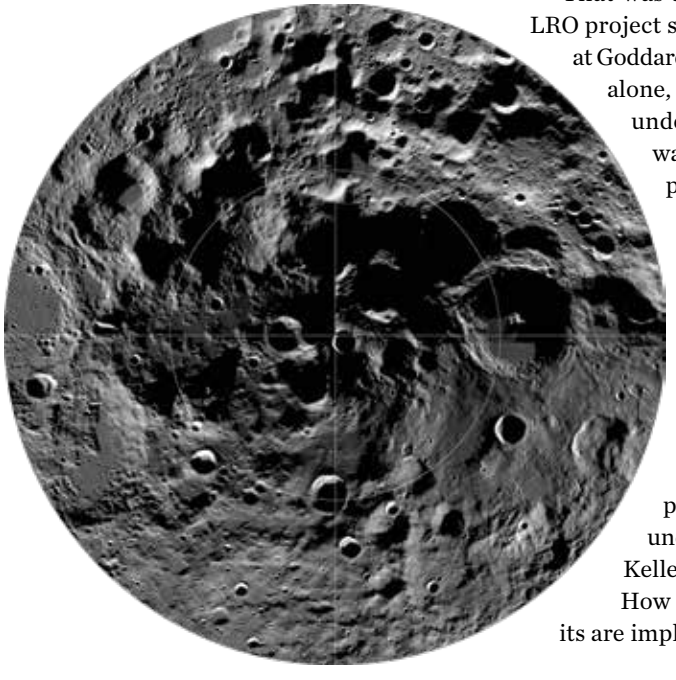
How the various water deposits are implanted and shuffled about is one enduring puzzle. How small subterranean pockets stayed warm for so long after the moon formed is another. Lava oozed on the surface in the last 100 million years, judging by smooth, dark terrains that are sparsely cratered. "This flies in the face with what was known about the moon," Petro says. "We thought lunar volcanism ended about a billion years ago."

Some changes are much more recent. In 2013, Earth-based telescopes detected a flash of light from the moon. LRO checked it out and found a new crater 18 meters across. "What was surprising was how far the ejecta went," Keller says. Debris had been tossed 35 kilometers — much farther than expected from an impact with a space rock estimated to be only about a meter wide.

Understanding what's currently hitting the moon and the traces those objects leave is crucial to interpreting the history of impacts plastered across the lunar surface. Similar impacts also affected Earth, but the signs of most have been erased by weather and geologic forces. "The moon is our way of studying the history of the Earth since the creation of the Earth-moon system," Petro says.

One of the seven instruments that LRO carries is a laser altimeter, a beam of light that scans and maps the surface in exquisite detail. "That's been a game changer," says Simone Marchi, a planetary scientist at the Southwest Research Institute in Boulder, Colo. "We can use the topography data to find old degraded craters that otherwise would not be easily detected in imagery."

Detailed maps reveal craters on top of other craters, laying out a rough sequence of when things hit the moon. Astronauts have brought back samples from some of these terrains, allowing researchers to use radiogenic dating to figure out when craters formed. That in turn supplies a record of what was smacking into other planets and asteroids. "We have a deep understanding of collisions going back to the beginning of the solar system," Marchi says. "That can only be done with the moon." ■



Water ice lurks within permanently shadowed craters at the moon's south pole, seen in this mosaic from the Lunar Reconnaissance Orbiter.

LRO/GSFC/NASA, ARIZONA STATE UNIV.

GENES & CELLS

How a Galápagos bird got tiny wings

Cormorant's flightlessness tied to several genetic changes

BY TINA HESMAN SAEY

Garbled signals from cellular antennas may have grounded the Galápagos cormorant.

Galápagos cormorants (*Phalacrocorax harrisi*) are the only cormorant species with wings too small to lift the birds off the ground. Broken primary cilia — antennas that cells need to receive developmental messages — left the birds with stunted wings, evolutionary biologist Alejandro Burga suggested May 12.

Burga, of UCLA, and colleagues compared DNA of flightless Galápagos cormorants with that of their close relatives, which have large wings and can fly. The researchers found more than 23,000 differences in more than 12,000 genes. Those changes have occurred within the last 2 million years, a short time by evolutionary standards.

The researchers narrowed down which genes might have had the biggest effect on cormorant evolution using a computer program that predicts whether a change in a gene will affect function. Of the genes predicted to have altered function, the researchers selected the 3.3 percent that have changed most drastically.

To determine what these genes do, Burga examined whether any of the human versions of these genes cause problems when they are mutated in people. Eight of the banged-up genes were associated with limb defects caused by faulty primary cilia, hairlike structures that grow from cells. The cilia receive signals important for cell development and functioning (*SN: 11/3/12, p. 16*). In people, genetic mutations that damage primary cilia lead to a variety of diseases, including developmental defects.

Normal versions of those eight genes are necessary for primary cilia to pick up signals sent by an important protein called hedgehog. Three other genes that are mutated in the flightless cormorants affect other aspects of the primary cilia.

It wasn't clear whether the cilia defects were the primary cause of the birds' flightlessness. So Burga focused on 10 of the altered Galápagos cormorant genes predicted by the computer program to give the biggest functional and evolutionary consequences. Those genes would be the most important wing shrinkers, Burga and colleagues reasoned.

One of those genes is called *CUX1*. The protein it produces helps turn on other genes. Most vertebrates have nearly identical versions of the gene. But in flightless cormorants, four amino acids have been lost from the protein, suggesting that it can no longer do its job or does it poorly. In chickens, a defective form of *CUX1* shrinks wings. Galápagos cormorant's altered form of *CUX1* might also make wings smaller because it fails to turn on limb growth genes.

Many researchers would have left the story at that, says evolutionary biologist Ludovic Orlando of the University of Copenhagen. But "they made an effort to validate their findings."

Burga and colleagues wondered whether *CUX1* and the primary cilia changes were related. The team injected cells used to mimic skeletal development in lab dishes with the normal vertebrate version of *CUX1*. Activity levels of two cilia genes rose by about 50 percent. That is evidence that *CUX1* normally helps to regulate activity of primary cilia genes.

But the Galápagos cormorant version of *CUX1* barely budged activity of the cilia genes. It also was not as good at stimulating growth and development of bone cells as the normal version, the researchers found. Those findings strengthen the case that *CUX1* and primary cilia together were involved in shrinking the flightless fowl's wings. ■

Galápagos cormorants' wings have dwindled so much over the last 2 million years that the birds can no longer fly. New genetic data implicate faulty cellular antennas, called primary cilia, in shrinking the wings.

BRIAN GRATWICKE/FLICKR (CC BY 2.0)

MEETING NOTE

Faulty gene can turn colds deadly for babies, toddlers

A faulty virus-sensing gene can make the common cold or respiratory syncytial virus deadly for babies and toddlers, a new study suggests.

Almost all children catch those viruses by age 2 or 3 years. Most kids quickly clear the viruses, but about one in 1,000 are admitted to the hospital intensive care unit with severe pneumonia. The reason some tykes get really sick is in their genes, Samira Asgari, a computational biologist at the Swiss Federal Institute of Technology in Lausanne, reported May 12.

Asgari and colleagues examined protein-coding DNA of 120 otherwise healthy toddlers and babies who ended up on respirators because of colds or RSV infections. Eight had one of three rare variants in the *IFIH1* gene that cause the gene's protein to be shorter than normal. That affects the protein's ability to detect double-stranded RNA made by some viruses and to turn on virus-fighting defenses. As a result, the viruses replicate better than normal.

Children who carry the variants have a harder time combating a first-time virus infection. If a child survives, the immune system learns to fight the virus by other means, Asgari said. The team also found mutations in other genes that may account for why other children get severely ill from common respiratory viruses. — *Tina Hesman Saey*

BODY & BRAIN

Crohn's genes block microbe messages

Normally, gut bacteria send signals that calm inflammation

BY MEGHAN ROSEN

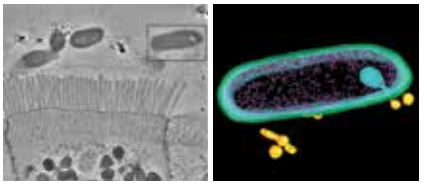
Good gut bacteria might not help people with Crohn's disease.

Protective microbial messages go unread in mice and human immune cells with certain defective genes, researchers report online May 5 in *Science*.

The findings are the first to tie together the roles of genes and beneficial microbes in the inflammatory bowel disease, says microbiologist Brett Finlay of the University of British Columbia in Vancouver, who was not involved in the new work.

"This is a major step forward in this area," he says. Human genes and friendly microbes work together to control inflammation, he says. "And when you muck that up, things can go awry."

In Crohn's disease, the immune system riles up too easily, triggering chronic inflammation. Researchers have linked the disease to glitches in nearly 200 genes, including *ATG16L1* and *NOD2*, which typically help kill bad bacteria.



A microbe (left, boxed) sends signals that calm the immune system in mouse intestinal cells (left, bottom). People with Crohn's disease may not get the messages, which are sent in pouches (right, yellow in microbe reconstruction).

Researchers have also reported that people with Crohn's have a different collection of gut microbes than healthy people do, says study coauthor Sarkis Mazmanian, a Caltech microbiologist. "There's a huge body of literature on the genome and on the microbiome," he says. But "no one knew what the interplay was between the two."

His team explored a potential link using a friendly gut microbe called *Bacteroides fragilis*, which sends out calming molecular messages that tell the immune system to tone down inflammation. Like letters inside envelopes, these messages travel in protective pouches called outer membrane vesicles, or OMVs.

Feeding message-filled OMVs to mice protects them from developing inflamed colons, or colitis — but not in mice lacking the Crohn's-linked genes *ATG16L1* and *NOD2*. When those mice receive a colitis-causing chemical, they succumbed to the disease, even after eating OMVs.

Mice without *ATG16L1* and *NOD2* genes "can't reap the benefits of the beneficial microbiota," Mazmanian says. Immune cells from human patients with defective versions of these genes didn't respond to OMVs either.

Mazmanian says finding ways to deliver *B. fragilis*' messages might help treat patients. Patients' cells dosed with just the contents of the OMVs (and not the protective pouch itself) actually got the message, his team finds. The treatment could have fewer side effects than other therapies, because it doesn't hamper the immune system, he says. ■

LIFE & EVOLUTION

Mitochondria go missing in microbe

Eukaryote breaks fundamental rule of complex life-forms

BY SUSAN MILIUS

A gut microbe collected from chinchilla droppings might be the first complex life-form to lack even a shred of a supposedly universal organelle.

Monocercomonoides, a one-celled gut microbe, apparently has no mitochondria, the organelles known as the cell's power plants. Cataloging DNA in the microbe turns up none of the known genes for mitochondrial proteins. Stealing genetic material from bacteria — which also survive without mitochondria — allowed the microbe to do without them, researchers report in the May 23 *Current Biology*.

Mitochondria are capsules that speckle the insides of all complex cells from pond scum to people, or so textbooks say. Some complex, or eukaryotic, cells look as if they have no mitochondria. So far, though, further searches have eventually detected mitochondrial remnants.

Monocercomonoides appears to have completely done away with mitochondria and the genes to make them, says study coauthor Anna Karnkowska, an evolutionary biologist now at the University of British Columbia in Vancouver.

This discovery marks "the most extreme mitochondrial reduction observed," says study coauthor Vladimír Hampl of Charles University in Prague.

The work also supports the idea that mitochondria lack a single core function. Although often described as cell powerhouses, mitochondria don't have much to do with supplying energy for cells living in low-oxygen or no-oxygen environments, Karnkowska says. For such cells, mitochondria can serve as more of a building studio. One supposedly essential mitochondrial function, scientists have said, is assembling clusters of iron and sulfur that activate useful cell compounds.

Bacteria and other simple, or prokaryotic, cells have their own assembly systems and don't need mitochondria to wall off the construction of iron-sulfur clusters. *Monocercomonoides* carries the genes for an assembly system that looks as if it was taken from bacteria.

The researchers made these discoveries while working out the DNA components that encode instructions for *Monocercomonoides*' proteins. There were no signs of chaperone proteins for conveying other proteins through mitochondrial membranes.

"Pretty amazing story," says Roland Lill of University of Marburg in Germany, who studies iron in cells. "The beauty of biology is that there are always amazing exceptions to basic biological rules." ■

MARK LADINSKY AND GREG DONALDSON/CALTECH

NANO for the Heart

Scientists are designing tiny 'missiles' to destroy waxy plaques in blood vessels

By Sarah C.P. Williams

Careening through the bloodstream, a single nanoparticle is dwarfed by red blood cells whizzing by that are 100 times larger. But when specially designed nanoparticles bump into an atherosclerotic plaque — a fatty clog narrowing a blood vessel — the tiny particles can play an outsized role. They can cling to the plaque and begin to break it down, clearing the path for those big blood cells to flow more easily and calming the angry inflammation in the vicinity.

By finding and busting apart plaques in the arteries, nanoparticles may offer a new, nonsurgical way to reduce a patient's risk for heart attack and stroke.

Nanoparticles measure less than 100 nanometers across — a thousandth the thickness of a dollar bill. Despite being tiny, they can be engineered to haul a mix of molecules — such as tags that make them stick to a plaque, drugs that block inflammation or dyes that let scientists track their movements. Over the last two decades, scientists have exploited these strategies to fight cancer, designing nanoparticles that deliver drugs (*SN Online: 1/3/14*) or dyes for imaging deep into the core of a tumor. The U.S. Food and Drug Administration has approved a few dozen cancer-focused nanomedicines.

Now researchers have begun engineering nanoparticles to target cardiovascular disease, which kills even more people each year than cancer. Nano-sized compounds have been built that can sweep into clogged arteries to shrink the plaques that threaten to block blood flow. Some nanoparticles home in on the plaques by binding to immune cells in the area, some do so by mimicking natural cholesterol molecules and others search for collagen exposed in damaged vessel walls. Once at the location of a plaque, either the nanoparticles themselves or a piggybacked drug can do the cleanup work.

The aim of all these approaches is to prevent strokes and heart attacks in people with cardiovascular disease, either before surgery becomes necessary or after surgery to prevent a second event. Today, cardiovascular nanoparticles are still far from pharmacy shelves. Most have not reached safety testing in patients. But in mice, rats and pigs, nanodrugs have slowed the growth of the plaques that build up on vessel walls, and in some cases have been able to shrink or clear them.

"I think the effect we can have with these nanoparticles on cardiovascular disease is even more pronounced and direct than what we've seen in cancer," says Prabhas Moghe, a biomedical engineer at Rutgers University in Piscataway, N.J.

Biological blockades

Every minute, more than a gallon of blood pumps through the human heart, pushing through miles of blood vessels to deliver oxygen and nutrients to organs and extremities. In a healthy person, the trip is as smooth as a drive on a freshly paved highway. But in the more than 10 percent of U.S. adults who have cardiovascular disease, the route might be more like a pothole-filled road squeezed by Jersey barriers.

Waxy globs, or plaques, of fat and cholesterol line the blood vessels, thickening and hardening the walls, impeding blood flow. As fat builds up inside the vessels, it also leaks into the vessel walls, swelling them and signaling the body to send immune cells to the area. The congregation of immune cells aggravates the blockage, the way emergency vehicles surrounding the site of a multicar pileup further slow traffic on a highway.

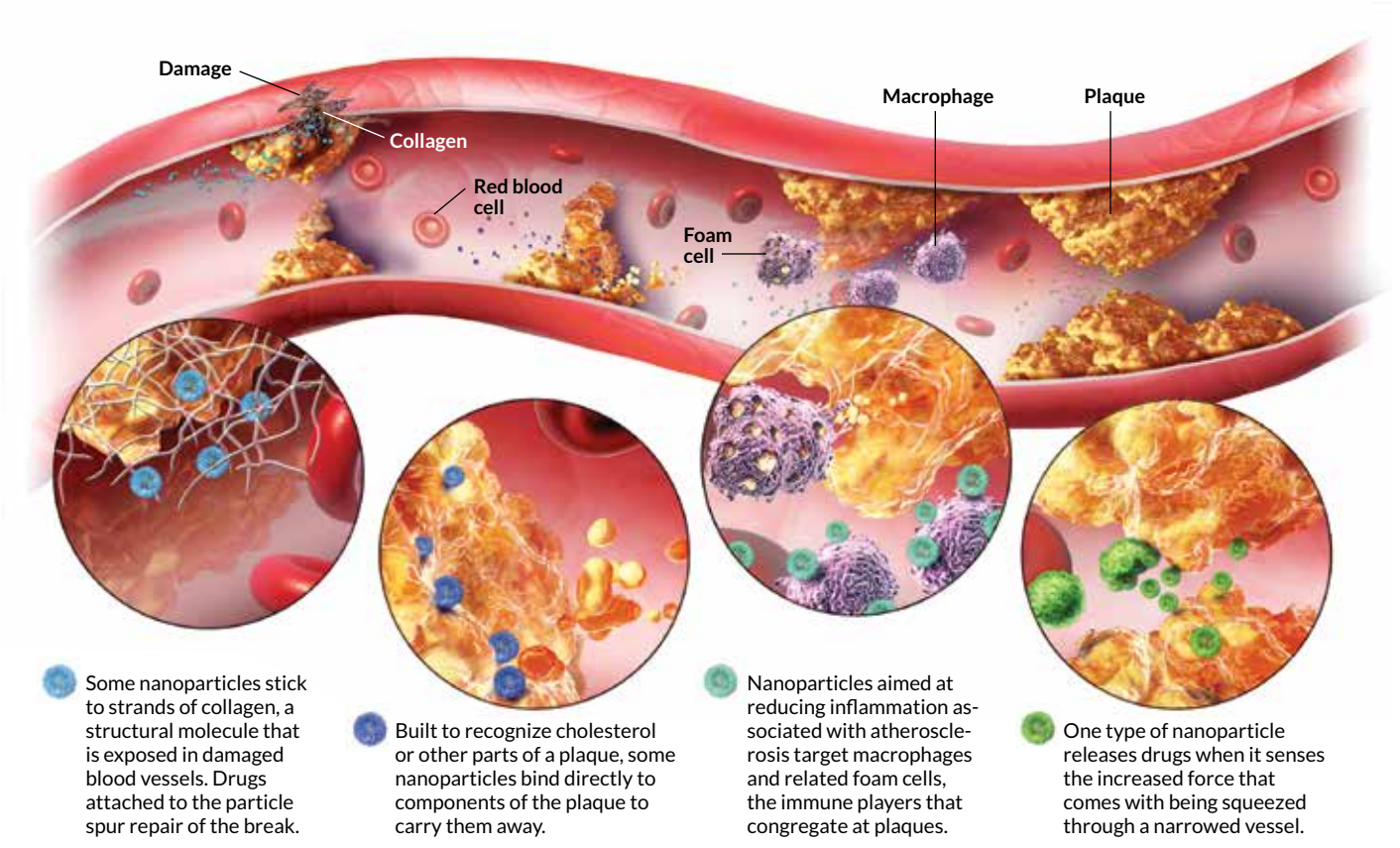
"The inflammation and the accumulation of fat in the walls of the blood vessel sort of feed off each other and exacerbate each other," Moghe says.

If the plaques grow large enough, or pieces chip off and travel to smaller vessels, they can block a vessel. If oxygen-filled blood can't reach the brain or heart, a stroke or heart attack results.

The drugs most often prescribed to prevent or treat atherosclerosis — plaque buildup on the inner walls of the arteries — are statins (*SN: 5/5/12, p. 30*). This class of drugs, available since 1987, slows the growth of the fatty plaques by lowering the amount of cholesterol circulating in the blood. But taking statins is akin to limiting the number of cars on a damaged road rather than

Drugs, dyes and targeting molecules can be arranged on the perimeter or inside of a nanoparticle (illustration shows one that mimics HDL).

S. MARRACHE AND S. DHAR/*PNAS* 2013



repairing potholes, some argue. And the drugs can boost a person's risk of diabetes and liver damage. In many cases, patients don't begin taking statins until they already have severe atherosclerosis, and the drugs do little to reverse the buildup of plaques that already exist.

"Heart disease is still the number one killer in the U.S.," says endocrinologist and biochemist Ira Tabas of Columbia University Medical Center. "So clearly this approach isn't working." Nanoparticles that can do what statins haven't been able to — shrink existing atherosclerotic plaques and eliminate the accompanying inflammation — could change that, Tabas and others say.

Macrophage magnet

To make nanoparticles congregate at the dangerous plaques, researchers need to identify something that makes the blockage stand out from the rest of the body. The crowds of immune cells near plaques act as a signpost that a plaque exists.

Many of the immune cells involved in atherosclerosis are macrophages, white blood cells that gulp pathogens, dead cells or debris in the body. At the site of a plaque, macrophages become swollen with fats and transform into what are called "foam cells" because of their foamy appearance. As they digest fats, foam cells send out chemical signals to recruit more inflammation-causing cells and molecules to the area. Because they're so intimately involved in the formation of plaques, macrophages and foam cells are a prime target for nanoparticles.

Moghe's group has designed nanoparticles that bind to molecules on the surface of macrophages, preventing them from gobbling fats and becoming foam cells. The researchers made the nanoparticles specifically target a subtype of macrophage that's involved in atherosclerosis, not the macrophages that might respond to other injuries in the body. When nanoparticles were injected into mice with narrowed arteries, the blockages decreased by 37 percent, Moghe's group reported last year in the *Proceedings of the National Academy of Sciences*.

Others are using cholesterol-like molecules as nanoparticle taxis to carry drugs to plaques and subdue the immune reaction. Statins aim to lower the form of cholesterol called low-density lipoprotein, which earned the name "bad cholesterol" for accumulating in plaques. High-density lipoprotein, or "good cholesterol," shuttles LDL away from these clogs to the liver, where it can be broken down. HDL also prevents macrophages from turning into foam cells and producing inflammatory molecules. So Shanta Dhar, a chemist at the University of Georgia in Athens, developed nanoparticles that mimic HDL. She presented the work in March in San Diego at a meeting of the American Chemical Society.

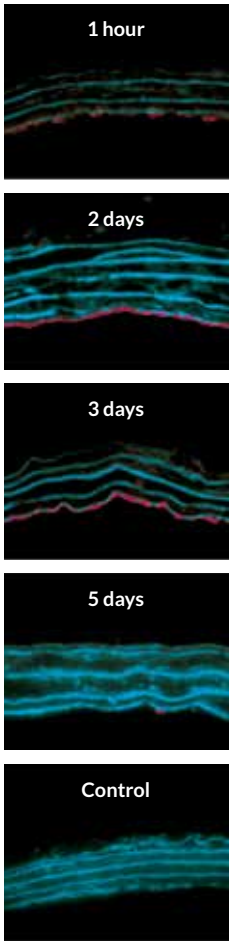
"HDL is our body's natural cholesterol-removing

Going places

To treat atherosclerotic plaques with nanoparticles, researchers have devised a variety of ways to send circulating particles directly to the fatty clogs. In each approach above, a molecule that's part of the nanoparticle binds to a molecule in or near the plaques.

SOURCES: M.E. LOBATTO *ET AL/ NAT. REV. DRUG DISCOV.* 2011; N. KORIN *ET AL/SCIENCE* 2012

NICOLLE RAGER FULLER



After injecting mice with nanofibers designed to bind to injured blood vessels, researchers tracked the location of the nanoparticles (red) and found that they bound to the inner layers of vessels (blue) within an hour and stayed put for three days.

nanomaterial," she says. In animal tests, the HDL-based nanoparticle can bind to free-floating macrophages circulating in the blood, just as HDL does, and follow them to a plaque, she explains. The nanoparticles can also bind to macrophages already glommed on to a plaque, and, mimicking the activities of natural HDL, carry the cells away.

Plaque buster

Willem Mulder, a nanomedicine researcher at the University of Amsterdam and the Icahn School of Medicine at Mount Sinai in New York City, has also designed HDL-mimicking nanoparticles. His particles deliver statins that make a beeline for macrophages and plaques, letting him administer the drug at lower-than-usual doses. He was inspired by earlier studies that showed how extremely high doses of statins, given to mice, could lower LDL levels while also packing anti-inflammatory properties. Of course, in humans, such high doses would probably cause liver or kidney damage. Mulder's solution: tack the statins to a nanoparticle to send them, missile-like, to the plaques. That way, a low dose of the drug could achieve the high concentration needed at the site of the atherosclerosis.

"We're exploiting the inherent targeting properties of HDL," he says. "And it works well with statins, which are small molecules."

In 2014 in *Nature Communications*, Mulder's group reported that plaque-filled arteries in mice given the nanoparticle were 16 percent more open than arteries in mice with no treatment, and 12 percent more open than in mice given a systemic statin. More work is needed to show whether these modest gains would translate to a reduced risk of heart attacks and strokes.

Others are using plaque-targeting nanoparticles to deliver anti-inflammatory drugs similar to methotrexate, which is used as a treatment for rheumatoid arthritis. The side effects of drugs like this, given systemically, are generally severe: vomiting, hair loss and "brain fog," to name a few.

"If someone with rheumatoid arthritis comes into your office completely crippled, it's worth all the side effects to put them on an anti-inflammatory drug," Tabas says. "But imagine someone with some risk factors for heart disease who feels great. They're not going to put up with these side effects."

Tabas thinks certain anti-inflammatories could be perfect candidates to tack on to nanoparticles because they would make possible lower doses with fewer side effects. He's awaiting the results of two large clinical trials testing non–nano-versions of the anti-inflammatory drugs methotrexate and anti-IL1 beta. It remains to be seen whether they're effective at clearing plaques and how severe the side effects are. If the drugs are effective, even with some side effects, Tabas says, it will give weight to his approach: activating the same pathways using targeted nanoparticles.

Tabas attaches his nanoparticles to a small section of a protein called annexin A1, which is involved in the same inflammatory pathway that many anti-inflammatory drugs target. His hope is that delivered only to an atherosclerotic plaque, the drug won't have the host of side effects that other immune blockers have.

Destination: vessel wall

The inflamed vessel wall around an atherosclerotic plaque goes through several changes in addition to the accumulation of belligerent immune molecules. As vessel walls are stretched and inflamed, the structural protein collagen, meant to keep the vessels taut and tubular, becomes exposed the way the threads of a tire begin to appear as it wears down. Scientists are using the exposed collagen to their advantage. Nanoparticles with a tag recognizing the collagen end up at plaques. But it's not as easy as affixing a GPS destination to the particles, says vascular surgeon Melina Kibbe of Northwestern University Feinberg School of Medicine in Chicago.

"It took us over a year of trying to find the right targeting [molecule] that would work," Kibbe says. Her nanoparticle combines a collagen-binding protein with nitric oxide, a molecule that stimulates the growth of new cells at wounds. To maximize the surface area of the drug that contacts the vessel wall, Kibbe's team arranged the molecules in a line, forming a nanofiber, rather than a sphere. As the fiber is swept through the bloodstream, it binds to exposed collagen, anchoring the nitric oxide in place to spur healing of the artery.

Kibbe and colleagues added fluorescent tags to the nanofibers and showed that the fibers congregated at injured spots on mouse arteries within an hour of injection. The tagged particles remained there for three days and the treated vessels ended up 41 percent more open, the researchers reported in the March *Antioxidants & Redox Signaling*.

Tabas also uses a collagen-binding protein, but his is organized in a more spherical shape to get his anti-inflammatory drugs to atherosclerotic plaques. In mice, the particles stayed in the plaques up to five days after treatment, shrinking the plaque by more than a third, his team reported

E.S.M. BAHNSON *ET AL/ANTIOXIDANTS & REDOX SIGNALING* 2016

in *Science Translational Medicine* in 2015. By comparison, some circulating statins last less than a day in the blood.

Rather than targeting proteins or immune cells, scientists at Harvard's Wyss Institute for Biologically Inspired Engineering have designed nanoparticles that are activated by the physical squeeze that comes with being swept through a narrowed artery. When the shear force around them increases, a cue that a plaque is present, the nanoparticles release their payload: a clot-dissolving drug called tissue plasminogen activator. The researchers reported late last year in *Stroke* that the nanoparticle, coupled with a stentlike device placed in the artery, increased the survival rate to more than 80 percent in mice that normally die of a clot entering their lungs.

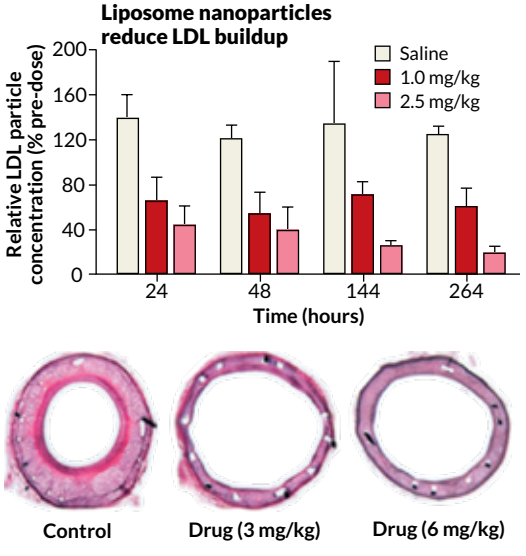
Pathway to patients

Nanoparticles currently in development for cardiovascular disease are still in animal testing. While no one has seen major side effects or toxicity in the animal trials so far, it remains a concern with a class of medicines that is so new.

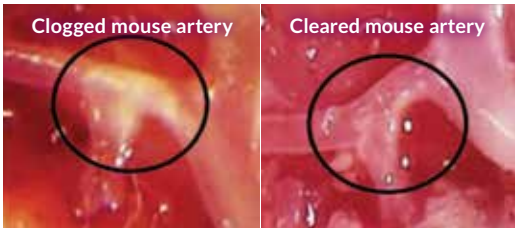
"We sometimes get so wrapped up in exuding only the good stuff about nanomedicine that we forget we also have to look at the side effects," Dhar says.

Another challenge for atherosclerosis drugs is determining who would benefit from treatment.

Clearing vessels Various plaque-targeting nanoparticles have been shown to be effective at lowering the levels of LDL in the blood of nonhuman primates (top graph) and at keeping rabbits' blood vessels open after a stent is inserted (bottom). The approach used in rabbits is now in early patient testing. SOURCE: M.E. LOBATTO *ET AL/NAT. REV. DRUG DISCOV.* 2011



FROM TOP: G. FREDMAN *ET AL/SCI. TRANSL. MED.* 2015; H.D. DANENBERG *ET AL/CIRCULATION* 2003



Mice fed high-fat diets had arteries clogged with yellow, fatty plaques. Animals treated with an anti-inflammatory nanoparticle had more effective plaque clearing (right) than did mice given a version of the nanoparticle without the drug (left).

Kibbe imagines her particles being used first in patients with severe atherosclerosis who receive stents or other invasive procedures to clear their plaques. The procedures are intended to help, she says, "but they actually are so traumatic that they cause injury to the vessel wall." Due in part to this renewed buildup in the arteries, people who have had one heart attack are at higher risk for a second. Even among people who have a permanent stent put in, which is designed to keep part of an artery clear, up to 20 percent become reblocked. Giving these patients nanoparticle-based drugs could keep them healthy, Kibbe says.

Taken to the next level, nanomedicines "certainly might be able to prevent plaques," she adds. Tabas imagines his nanoparticles given as a once-a-month injection, but that's speculation.

Moving to test nanoparticles as a preventive — in the huge percentage of the population at risk for atherosclerosis — is probably a long way off, Mulder says. According to the U.S. Centers for Disease Control and Prevention, around half of all adult Americans have one of the top risk factors for cardiovascular disease.

"I really don't foresee that you would start preventively treating patients who don't have symptoms with nanoparticles," Mulder says. "But to take a person who's hospitalized after a heart attack and stick a needle in their arm and infuse nanoparticles, that's not hard."

Once a few drugs have been validated as working in clinical trials, researchers expect progress to speed up, since the drug cargo on a nanoparticle engineered to target a plaque could easily be switched out for other drugs. Designing the particles, says Moghe, "is almost like building with pieces of Lego." ■

Explore more

- Mark E. Lobatto *et al.* "Perspectives and opportunities for nanomedicine in the management of atherosclerosis." *Nature Reviews Drug Discovery*. November 2011.

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