Children are naturally inquisitive. “Mom. Mom. Mom. Why is the sky blue?” and all that sort of stuff. Sung Chi Tuan was no different. As a tyke growing up in Hong Kong, he was full of questions. Often he'd take it upon himself to find the answers.

His father had a few wristwatches. On each was engraved the number of jewels contained in each timepiece. In watch parlance, a jewel is a little pivot used to reduce friction between all the moving parts that help keep time.

Young Master Tuan did not know this. “So I would take the watches apart to find the jewels,” Tuan says with a hint of decades-long disappointment tinting his words.
“Remember early attempts at building airplanes? They tried to make them look like birds, but mechanically it didn’t work. When the Wright Brothers built their airplane, it was totally different from the way a bird flies. It’s got wings, but it’s nothing like a bird.”
“I’ve become very interested in repairing, regenerating, and engineering new tissues, particularly [those] related to the skeleton, and it all started with looking at how this calcium business works,” he says.

Lo says her husband’s tendency to yield easily to curiosity and grasp opportunity—as manifested in the Aristotle, egg, chicken, and calcium transport equation—was evident even when he was an undergrad. “He loves music, which he didn’t have the opportunity to study when he was growing up,” she says. “When he went to Berea College, he was in work study at the music library and listened to all kinds of music. He got his own musical education. His taking advantage of chances is a character trait.”

That spell as a musical autodidact led to Tuan performing with the Tanglewood Festival Chorus in Boston. “He has a nice baritone voice,” Lo says. Will he do the same with the Pittsburgh Symphony’s Mendelssohn Choir? “Maybe. He hasn’t exactly had a lot of time as of late,” she notes.

As Tuan gets his Pittsburgh lab up to speed, he’s no longer particularly interested in chickens; he’s more concerned with how to create cartilage, tendon, and ligament tissues to replace those ravaged by injury or age. When these tissues are injured or worn by overuse, the result is often osteoarthritis—a painful, chronic, and difficult-to-treat condition that affects some 27 million Americans, according to the NIAMS.

At present, osteoarthritis is managed through pain medication, physical therapy, and, in severe cases, surgery to repair the damaged tissue or replace joints. The problem with surgery is that the mended tissue—scar tissue, essentially—cannot bear the same weight as the native, healthy tissue. Further, it has a tendency to erode over time, necessitating follow-up procedures. Replacement joints, as sophisticated as they have become, don’t hold up well over time.

Tuan says we can do better, and we can do so by mimicking nature.

Making man-made, or rather, “man-assisted” tissue in vitro from adult stem cells, particularly mesenchymal stem cells that are drawn from bone marrow, muscle, or fat, is the easy part—not that it’s particularly easy. The real difficulty arises, Tuan says, in making something that looks like muscle, cartilage, or a spinal disc function like muscle, cartilage, or a spinal disc.

“The mechanical properties are lousy, but it’s a beginning,” Tuan told Wired magazine in 2008 of his earlier efforts.

To improve function and make the new tissue as good as the old, Tuan has decided to give these stem cells something of a kick-start. With knowledge about how cells differentiate during embryonic development—what growth factors play which roles at what times—Tuan is capable of turning mesenchymal stem cells into what he wants, or at least a fair simulacrum of it. But this is clearly not enough.

“This is a little different from the classical engineering approach of ‘Oh, something’s missing, I’ll just build something that looks like it,’” he says. “In the body, it doesn’t quite work that way. Remember early attempts at building airplanes? They tried to make them look like birds, but mechanically it didn’t work. When the Wright Brothers built their airplane, it was totally different from the way a bird flies. It’s got wings, but it’s nothing like a bird.”

Tuan’s challenge, however, is a bit different from what Orville and Wilbur Wright faced as they, in effect, cheated their way around the avian model to achieve flight. Tuan has got to build tissue that not only looks like what it’s intended to replace but also essentially is what it’s intended to replace.

To ease the transition from stem cell to tissue, Tuan decided to build a template. In normal human development, embryonic stem cells, as they differentiate, assemble themselves into heart, bone, muscle, cartilage, etc. This is easier for them because, as Tuan says, embryonic stem cells are much more adept at becoming whatever they need to become. Adult stem cells are less malleable, but still open to influence and less likely to proliferate out of control.

“An embryonic stem cell is like a kindergartner—it doesn’t know very much, but it can become anybody as long as it’s taught the right things,” Tuan says. “Adult stem cells are more like high school graduates. They’ve already learned a few things but have developed some bad habits. Still, though, you can influence them by putting them with the right friends.”

Their best friend, in Tuan’s metaphor, is a scaffold matrix. Without a space-filling guide the mesenchymal stem cells can glom onto, there’s a high risk that they’ll be swept away before they can become tissue. “Unless you can make cells happy,” Tuan adds, “you’re not going to have the right kind of tissue in the end. What do cells like? Well, they live in a matrix that they make themselves.” Logic then dictated to Tuan that he try to make a matrix as close in appearance, structure, and function to the original matrix as he could.

In high-resolution microscopy, an “original matrix” appears as a bunch of nanometer-
scale fibers. Tuan takes a liquid polymer and, using a technique borrowed from the textile industry, spins the stuff rapidly in the presence of a strong electrical field. As the polymer attempts to diffuse the charge, it forms into—you guessed it—a bunch of nanometer-scale fibers.

“The cells love this!” Tuan says. “They think they’re at home. It’s assisted development. Otherwise it might take them a while to put all that stuff [the matrix] together. Now that we gave them the stuff, they can decorate it with their own molecules.”

The scaffold can also be treated with other molecules that help attract mesenchymal stem cells and induce them into turning into what Tuan wants them to become. “We can attach those [biologically active] reactive groups to the fibers according to whatever we want,” Tuan says. “What we have done now is load these fibers with very small molecules, and we can add hormones or growth factors. Let’s say you have osteoarthritis; you can add stem cells at up to 4,000 times the density found in bone marrow and much, much higher than found in normal muscle tissue. This overzealous attempt at healing is likely what causes bone to form, but Tuan has yet to sort out why the stem cells make this error in differentiation.

Some good, though, has come out of this problem. These extra stem cells, Tuan found, also have the ability to induce nerve growth. He suggests that perhaps these cells can be harnessed to induce peripheral nerve repair. “We’re not too far along on this,” Tuan says, “but it’s a work in progress for sure.”

Tuan’s not alone in Pitt orthopaedics when it comes to regenerating tissue, particularly cartilage. His colleague Constance Chu mines a similar vein. Chu, the Albert Ferguson Associate Professor of Orthopaedic Surgery and director of the Cartilage Restoration Center at Pitt, is preparing to test her work on horses.

An MD, Chu recently received a $1.7 mil-

lion Grand Opportunities grant from the NIH for her project “Multicenter Cartilage Repair Preclinical Trial in Horses.” She’s collaborating with investigators from Cornell University, Colorado State University, and the University of California at San Diego.

Chu also works with bioactive scaffolding and bone marrow stem cells, but her Grand Opportunities grant takes another approach: “What I’m doing I call ‘back-table tissue engineering.’ Basically, we’re aspirating bone marrow from horses and concentrating stem cells and clotting factors by centrifuge in the operating room.” This material can lead to tissue regeneration once implanted in the knee.

While Chu admires, and shares, Tuan’s approach, she thinks lab-engineered cartilage may take longer to reach the clinic than her “back-table” work. “With gene therapy or nanotechnology or synthetic polymers, there are major hurdles regarding proving safety and efficacy,” she says.

If the horse trial goes well, Chu plans to pursue clinical trials in humans within two years.

Tuan sees a lot of promise in Chu’s work. In the meantime, he’s going to keep busy with his scaffolds. “When you’re dealing with tissues subjected to the pressures that go along with being in a ligament or tendon.”

Freddie Fu, David Silver Professor and chair of the Department of Orthopaedic Surgery at Pitt is confident his new recruit will find the answers he seeks. “I’ve been following his career for 20 or 25 years now, even before he was at the NIH, and I think he’s leading the world in this biological approach to the problem,” Fu says. “It’s a solvable problem, and I think he’s going to find a solution.”

Tuan says he’s glad to be at Pitt to pursue this stage of his work.

“For the kind of work I am interested in, Pittsburgh is a powerhouse,” Tuan says. “I don’t have to come in here and build anything, which is actually nice. Here, science is truly multidisciplinary, and I think it’s a lot more fun when you have different kinds of people thinking about these problems. It’s great to be in a place with that kind of interaction.”

Well, Tuan concedes (as construction workers drill and hammer nearby), he does have to build something—cartilage, muscle, tendon. After all, that’s why he uprooted his lab and came to the banks of the Monongahela River.