Good as new? NBA star Greg Oden (*left*) underwent surgery last year to rebuild cartilage in his knee.

TISSUE ENGINEERING

Coming Soon to a Knee Near You: Cartilage Like Your Very Own

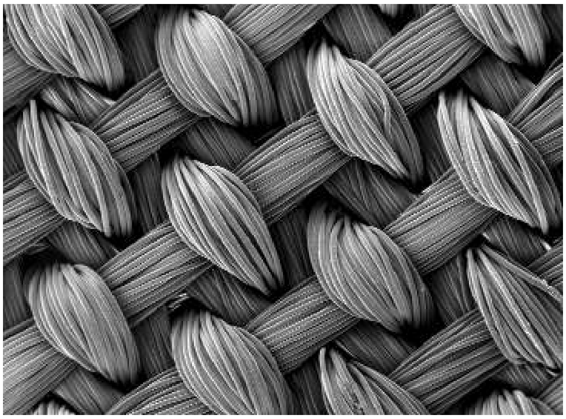
Weaving materials science and biology together, researchers are drawing closer to the elusive goal of recreating tissues that do the body's work, such as cartilage and muscle

BASKETBALL STAR GREG ODEN HASN'T MET Farshid Guilak, a biomedical engineer at Duke University in Durham, North Carolina. But as an insurance policy, Oden may want to send Guilak a few tickets each time his Portland Trailblazers play in nearby Charlotte. Oden was the first player selected in the 2007 National Basketball Association draft, yet he was sidelined his entire rookie season because of a bad right knee. After cleaning out damaged cartilage in the knee, doctors performed microfracture surgery, punching several tiny holes at the tip of the surrounding bones so that the resulting influx of blood would ferry bone marrow stem cells into the region; those cells, they hoped, would later differentiate into cartilage-producing chondrocytes and repair Oden's knee.

Now, several games into this year's season, Oden's knee seems as solid as his monster dunks. But microfracture surgeries don't always help athletes recover. And they can't aid the millions of people with bad knees or hips due to more widespread cartilage problems, such as arthritis sufferers.

Guilak has a potential solution: Engineer new cartilage by seeding chondrocytes onto an ultrastrong, woven polymer matrix and implant that matrix into patients. His tissue-engineered cartilage isn't ready for human trials yet, so patients like Oden should be careful for now. Still, it's getting close, and Guilak's strategy is a prime example of how materials scientists are teaming up with biologists to engineer tissues that perform mechanical work, such as cartilage, ligaments, and even heart muscle.

Recreating such mechanically active tissues was long thought to be easier than artificially building complex organs such as the liver and pancreas. (See special section on how organs naturally form, p. 1489.) But imitating the mechanical feats performed by natural tissue is a tall order. For example, the cartilage in your body's joints can withstand 12 megapascals of pressure—more than 10 times the amount generated if you hang from a ledge by a single fingernail. Researchers have already made significant progress in building new bone that's as strong as the natural stuff (*Science*, 1 September 2000, p. 1498). Now, they are extending that success to softer mechanically active tissues by learning to mimic many of their key attributes. "It's a breakthrough time in this field, where we are seeing several biomimetic capabilities coming together," says Guilak.



Next best thing. A woven scaffold made of a biodegradable polymer helps seed cells that can produce new cartilage.

Crunched cartilage

Off the basketball court, the most common type of cartilage damage is the widespread loss of tissue that's a hallmark of many forms of arthritis. According to the Arthritis Foundation, arthritis costs the U.S. economy alone $128 billion per year in medical bills and indirect expenses, including lost wages and productivity. In arthritis patients, the healthy cartilage that lubricates and cushions the impact between adjoining bones breaks down over time. Bones then rub directly against one another, causing pain and loss of movement in the joint. Cartilage contains no nerves. So by the time patients feel pain, significant amounts of cartilage may already be gone.

When the loss is slight, as was the case with Oden, doctors use microfracture surgery to trigger new cartilage growth, even though replacement cartilage is typically weaker than the original. Other treatments include transplanting cartilage from elsewhere in the patient's body into the afflicted area, as well as harvesting a person's chondrocytes from a region of healthy cartilage, amplifying them in a lab, and reinjecting them into a patient's joint. But none of these efforts produce cartilage that's as strong as the original. And when cartilage loss is extensive, doctors can do little other than completely replace the joints with metal and plastic prostheses. That can offer immediate pain relief, but replacement joints often wear out after only a decade and can typically be replaced only once, due to accumulated damage to the patient's bones, says Gerard Ateshian, a biomedical engineer at Columbia University.

As an alternative, cell-free synthetic cartilage has made some progress. In select cases, doctors can inject a polymer gel into joints to help ease symptoms. But such procedures often offer only temporary relief because the gel breaks down. Prospects are a bit better for gels used to replace damaged disks between vertebrae in the spine; those gels can be injected and contained within the thin, fibrous sac that houses the cartilagelike material of a normal disk. Michele Marcolongo and colleagues at Drexel University in

CREDITS (TOP TO BOTTOM): LUCY NICHOLSON/REUTERS/LANDOV; FRANK MOUTOS AND FARSHID GUILAK

Philadelphia, Pennsylvania, for example, have made gels from a copolymer of polyvinyl alcohol and polyvinyl pyrrolidone. The gels are tough and resilient, capable of withstanding up to 10 million cycles of compression and release in lab studies. Marcolongo's technology has been picked up by the medical devices company Synthes Spine, whose preclinical studies on animals suggest that the treatment seems to "completely restore mechanics," Marcolongo says.

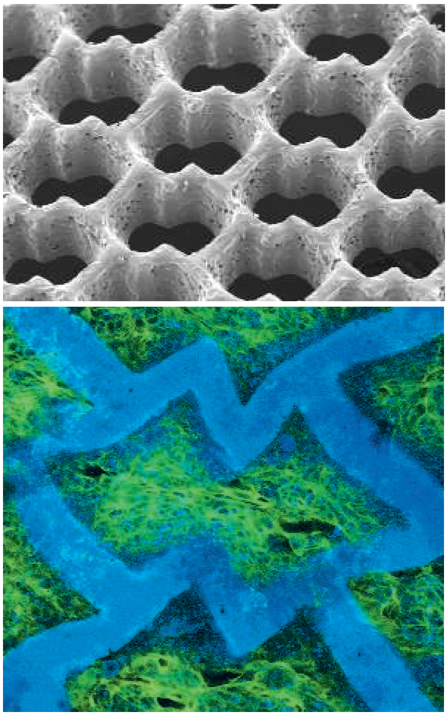
Fully synthetic gels have had less success in the knee and other less well-confined spaces. The arrangement of these joints requires that the cushioning material in the middle be able to withstand as much as 10 times a person's weight. Gels compressed with that much force typically squish out to the sides, like a spoon pressing down on a slab of Jell-O. By contrast, collagen fibers in natural cartilage make it extremely stiff so that it resists squishing outward under an applied force, Ateshian says.

Stronger scaffolds

To craft such strong tissue, researchers have focused on coaxing the body to grow its own additional cartilage cells on a synthetic template, rather than trying to recreate cartilage from scratch. Researchers have seeded cartilage-producing chondrocytes onto synthetic scaffolds in vitro for decades in hopes that this would cause the cells to generate new cartilage with the same impressive properties as the native version. But the results have almost always been disappointing. Chondrocytes do grow and put out a mixture of collagen and charged compounds called proteoglycans. But the resulting cartilage winds up far weaker.

Ateshian has recently made tougher cartilage by applying a little force of his own. Growing cartilage can sense mechanical stress and responds by becoming stronger, akin to the way that weight training helps build strong bones. Ateshian applied this principle back in 2000, when his team reported seeding a culture of chondrocytes onto a synthetic hydrogel and compressing the gel in a chamber. The resulting new cartilage was five times stronger than that created without mechanical loading. Recently, the team has boosted that strength up to about 20% of that of native cartilage by cycling the compression on and off and adding a cocktail of growth factors.

In a variation on this theme, Rocky Tuan, a tissue engineer at the National Institute of Arthritis and Musculoskeletal and Skin Diseases in Bethesda, Maryland, is also putting weight on tissues and adding growth factors. But Tuan and his colleagues deposit their cells atop fibers of a biodegradable polymer called poly(α-hydroxy ester). Tuan first learned to spin the fibers a decade ago with an apparatus akin to those that spin cotton candy from sugar. His lab has since perfected techniques to align the fibers to better control cartilage growth and resist compression. Tuan says his team's artificial cartilage now also has about 20% of the strength of native cartilage. "We would like to get to 40% to 50%," Tuan says. He adds that most clinicians believe that will be good enough to restore mobility for many patients. In a paper in press at the *Journal of Tissue Engineering and Regenerative Medicine*, Tuan and his colleagues report that after implanting their synthetic cartilage into pigs' hip joints, the material seemed to integrate well with the native cartilage; the animals appeared to walk normally as well.



Mending broken hearts. A polymer scaffold (*top and above in blue*) causes heart muscle cells (*above in green*) to align and contract in a preferred direction.

Tuan's nanofiber scaffolds, however, have very small pores, making it difficult for chondrocytes to penetrate and churn out new cartilage. Guilak's team has made progress with a scaffold that leaves plenty of room for the cells. He, Franklin Moutos of Duke, and Lisa Freed of the Massachusetts Institute of Technology (MIT) in Cambridge have created a novel three-dimensional weaving technique for building high-strength scaffolds. The trio wove their scaffold with a yarn made from a biodegradable polymer called polyglycolic acid (PGA) and seeded it with chondrocytes, they reported in *Nature Materials* last year. The woven fabric gave the scaffold compressive, tensile, and sheer strength on the same order of magnitude as native cartilage. That's a "major advance" and "an exciting opportunity for tissue engineers," Ateshian wrote in a commentary at the time.

Guilak's engineered cartilage still had some drawbacks. The PGA used to weave the scaffold degraded in about 2 weeks, too quickly for the seeded chondrocytes to churn out the collagen and proteoglycans needed to rebuild strong cartilage. Guilak's team has since turned to another polymer known as poly(ε-caprolactone) that degrades more slowly. Cytex Therapeutics, a biotech start-up in Durham, North Carolina, is now carrying out preclinical animal studies with the artificial cartilage and hopes to launch human trials in 2010.

Freed and her MIT colleagues have also been working to extend their success with patterned scaffolds to other tissues as well. Freed and postdoc George Engelmayr Jr. recently led a team that created artificial heart-muscle tissue from a honeycomb-shaped scaffold that flexes like an accordion. Cells in heart muscle, Freed explains, are aligned in specific directions to coordinate how the muscle flexes. Most biomaterial scaffolds, however, can't reproduce this alignment.

To do so, the team used a computer-controlled excimer laser to cut a precise pattern of holes and grooves in successive layers of a rubberlike degradable polymer scaffold. They then seeded the scaffold with neonatal rat heart cells. Not only did the cells grow in a preferred orientation, but when prompted by an electric field, they also contracted in the preferred direction, the team reports in the December issue of *Nature Materials*.

The group is working to optimize the pores in the structure and to build a microfluidic perfusion chamber to feed nutrients into the cells. Ultimately, Freed adds, the hope is that the novel scaffolds can be used to create artificial heart-muscle patches to repair small sections of diseased heart tissue. Guilak says he's impressed with Freed's team's ability to pattern cells: "You could apply this to a number of tissues, including tendons and ligaments." Tissue engineers are hoping not only that this will happen but also that the results will begin to alleviate the suffering of arthritis patients and perhaps even keep a few basketball stars on the court.

–ROBERT F. SERVICE

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